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16. ABSTRACT
Inorganic arsenic, predominantly the tri- and pentavalent forms, is emitted to the environment primarily through smelting activity, biocide use and glass manufacturing. Monitoring data indicate a concentration of equal to or less than 0.1 $\mu g/m^3$ for most locations. Major routes of absorption of inorganic arsenic in the general population are inhalation and ingestion. Inhaled inorganic arsenic deposited in the lungs is eventually absorbed. Most ingested soluble inorganic arsenic is absorbed, whereas insoluble forms pass through the gastrointestinal tract with negligible absorption. Inorganic arsenic metabolism in man is complicated by biotransformation processes which include the methylation and oxidation-reduction interconversion of inorganic arsenic. Long-term accumulation of inorganic arsenic does not generally occur in physiologically active compartments in the body; renal clearance appears to be the major route of excretion of absorbed inorganic arsenic. Acute symptoms of inorganic arsenic poisoning include severe gastrointestinal damage, facial edema, cardiovascular reactions, peripheral nervous system disturbances, and hematopoietic system effects. General population concerns arising from long-term exposures to moderate levels of inorganic arsenic include respiratory tract cancer, skin cancer, non-cancerous skin lesions, peripheral neuropathological effects and cardiovascular effects. There appears to be a nutritional requirement for low levels of inorganic arsenic in certain experimental animals; however, this requirement has not yet been established in man.

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Office of Health and Environmental Assessment
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This document has been reviewed in accordance with the U.S. Environmental Protection Agency's peer and administrative review policies and approved for presentation and publication. Mention of trade names or commercial products does not constitute endorsement or recommendation for use.

PREFACE

The Office of Health and Environmental Assessment, in consultation with other Agency and non-Agency scientists, has prepared this health assessment to serve as a "source document" for Agency-wide use. Specifically, this document was prepared at the request of the Office of Air Quality Planning and Standards.

In the development of this assessment document, the scientific literature has been inventoried, key studies have been evaluated, and summary/conclusions have been prepared such that the toxicity of arsenic is qualitatively and, where possible, quantitatively identified. Observed effect levels and dose-response relationships are discussed where appropriate in order to place adverse health responses in perspective with observed environmental levels.

ABSTRACT

This document summarizes current scientific information regarding the effects of inorganic arsenic on man and the environment. The observed effects, as presented herein, constitute the health basis from which the U.S. Environmental Protection Agency will make determinations regarding regulatory initiatives pursuant to the Clean Air Act. Specifically, this document discusses the following topics--physical and chemical properties of inorganic arsenic; environmental distribution and cycling; inorganic arsenic metabolism; toxicology, including mutagenic and carcinogenic properties; and essentiality--and identifies the factors most germane to assessing quantitative and qualitative human health risks.

TABLE OF CONTENTS

Page

LIST OF TABLES.. ix
LIST OF FIGURES... xi

1. INTRODUCTION... 1-1

2. SUMMARY AND CONCLUSIONS.. 2-1
  2.1 BACKGROUND INFORMATION.. 2-1
    2.1.1 Chemical/Physical Aspects of Arsenic.......................... 2-1
    2.1.2 The Environmental Cycling of Arsenic.......................... 2-3
    2.1.3 Levels of Arsenic in Various Media............................ 2-4
      2.1.3.1 Levels of Arsenic in Ambient Air.......................... 2-4
      2.1.3.2 Levels of Arsenic in Drinking Water....................... 2-4
      2.1.3.3 Arsenic in Food... 2-5
      2.1.3.4 Arsenic in Soils.. 2-5
      2.1.3.5 Other Sources of Arsenic.................................. 2-5
  2.2 ARSENIC METABOLISM.. 2-6
    2.2.1 Routes of Absorption.. 2-6
      2.2.1.1 Respiratory Absorption.................................... 2-6
      2.2.1.2 Gastrointestinal Absorption............................... 2-6
      2.2.1.3 Transplacental Transfer................................... 2-6
    2.2.2 Biotransformation of Inorganic Arsenic In Vivo................ 2-7
    2.2.3 Distribution of Arsenic in Man and Animals.................... 2-8
    2.2.4 Arsenic Accumulation.. 2-9
    2.2.5 Arsenic Excretion... 2-10
  2.3 ARSENIC TOXICOLOGY.. 2-10
    2.3.1 Acute Toxicity.. 2-10
    2.3.2 Chronic Toxicity.. 2-11
      2.3.2.1 Mutagenesis of Inorganic Arsenic.......................... 2-11
      2.3.2.2 Carcinogenesis of Inorganic Arsenic....................... 2-12
        2.3.2.2.1 Human Epidemiology of Arsenic Carcinogenesis.......... 2-13
        2.3.2.2.2 Experimental Studies of Arsenic Carcinogenesis........ 2-18
      2.3.2.3 Chronic Neurological Effects of Arsenic Exposure.......... 2-19
      2.3.2.4 Cardiovascular Effects of Arsenic Exposure................ 2-20
      2.3.2.5 Other Systemic Effects of Arsenic......................... 2-20
    2.3.3 Factors Affecting Arsenic Toxicity............................ 2-21
  2.4 ARSENIC AS AN ESSENTIAL ELEMENT................................... 2-21
  2.5 HUMAN HEALTH RISK ASSESSMENT FOR ARSENIC.......................... 2-22
    2.5.1 Exposure Aspects of Arsenic................................... 2-22
    2.5.2 Effect/Response Aspects of Arsenic............................ 2-23
      2.5.2.1 Relevant Health Effects................................... 2-23
      2.5.2.2 Dose-Effect/Dose-Response Relationships................... 2-24
    2.5.3 Populations at Special Risk to Health Effects of Arsenic...... 2-27

TABLE OF CONTENTS
(continued)

Page

3. BACKGROUND INFORMATION.......... 3-1
  3.1 CHEMICAL/PHYSICOCHEMICAL ASPECTS.......... 3-1
  3.2 ENVIRONMENTAL CYCLING OF ARSENIC.......... 3-4
  3.3 LEVELS OF ARSENIC IN VARIOUS MEDIA.......... 3-8
    3.3.1 Levels of Arsenic in Ambient Air.......... 3-9
    3.3.2 Levels of Arsenic in Drinking Water.......... 3-15
    3.3.3 Arsenic in Food.......... 3-16
    3.3.4 Arsenic in Soils.......... 3-19
    3.3.5 Other Sources of Arsenic.......... 3-20

4. ARSENIC METABOLISM.......... 4-1
  4.1 ROUTES OF ARSENIC ABSORPTION.......... 4-1
    4.1.1 Respiratory Absorption.......... 4-1
    4.1.2 Gastrointestinal Absorption.......... 4-6
    4.1.3 Transplacental Passage.......... 4-10
  4.2 BIOTRANSFORMATION PROCESSES IN VIVO.......... 4-11
    4.2.1 Biomethylation of Inorganic Arsenic in Humans and Experimental Animals.......... 4-11
      4.2.1.1 Human Studies.......... 4-12
      4.2.1.2 Animal Studies.......... 4-15
    4.2.2 *In Vivo* Oxidation/Reduction of Inorganic Arsenic in Mammalian Systems.......... 4-15
    4.2.3 Chemical Stability of Trivalent and Pentavalent Inorganic Arsenic to Oxidation-Reduction.......... 4-20
  4.3 DISTRIBUTION OF ARSENIC IN MAN AND ANIMALS.......... 4-20
  4.4 ARSENIC ACCUMULATION.......... 4-23
  4.5 ARSENIC EXCRETION IN MAN AND ANIMALS.......... 4-24

5. ARSENIC TOXICOLOGY.......... 5-1
  5.1 ACUTE TOXICITY OF ARSENIC IN MAN AND ANIMALS.......... 5-1
  5.2 CHRONIC TOXICITY OF ARSENIC IN MAN AND ANIMALS.......... 5-2
    5.2.1 Neurotoxic Effects.......... 5-2
    5.2.2 Cardiovascular Effects.......... 5-8
    5.2.3 Teratogenesis and Developmental Effects.......... 5-11
      5.2.3.1 Animal Studies.......... 5-11
      5.2.3.2 Human Studies.......... 5-16
    5.2.4 Hematological Effects.......... 5-17
    5.2.5 Hepatic Effects.......... 5-18
    5.2.6 Renal Effects.......... 5-19
    5.2.7 Respiratory Effects Other Than Cancer.......... 5-20
    5.2.8 Immunosuppressant Effects.......... 5-20
  5.3 FACTORS AFFECTING ARSENIC TOXICITY.......... 5-21

6. ARSENIC MUTAGENICITY.......... 6-1
  6.1 GENE MUTATION STUDIES.......... 6-1
    6.1.1 Prokaryotic Test Systems (Bacteria).......... 6-1
    6.1.2 Eukaryotic Microorganisms (Yeast, Fungi).......... 6-7
    6.1.3 Mammalian Cells *In Vitro*.......... 6-7
  6.2 CHROMOSOMAL ABERRATION STUDIES.......... 6-8

TABLE OF CONTENTS
(continued)

Page

6.2.1 Chromosomal Aberrations In Vitro.................... 6-8
6.2.2 Chromosomal Aberrations In Vivo..................... 6-15
6.2.3 Chromosomal Aberrations and SCE in Cells of Arsenic-Exposed Human Beings................................ 6-16
6.3 OTHER STUDIES INDICATIVE OF MUTAGENIC DAMAGE............... 6-23
6.3.1 Preferential Killing of Repair-Deficient Bacterial Strains.. 6-23
6.3.2 DNA Damage in Mammalian Cells In Vitro............... 6-30
6.3.3 Fidelity of DNA Synthesis.......................... 6-30
6.3.4 Induction of Sister Chromatid Exchanges (SCE) In Vitro 6-31
6.4 INTERACTIONS OF ARSENIC WITH OTHER MUTAGENS................ 6-32
6.4.1 Bacterial Studies.................................. 6-32
6.4.2 Mammalian Cells In Vitro........................... 6-37
6.4.3 Mammalian Cells In Vivo............................ 6-37
6.5 POSSIBLE MECHANISMS OF ACTION............................. 6-39

7. ARSENIC CARCINOGENICITY..................................... 7-1
7.1 HUMAN STUDIES... 7-1
7.1.1 Clinical Aspects of Human Carcinogenesis............ 7-1
7.1.2 Epidemiological Aspects of Human Arsenic Carcinogenesis..................................... 7-12
7.1.2.1 Cancer of the Lung.......................... 7-12
7.1.2.2 Cancer of the Skin and Precancerous Skin Lesions..................................... 7-52
7.1.2.3 Other Cancers............................... 7-74
7.2 ANIMAL STUDIES.. 7-77
7.3 QUANTITATIVE CARCINOGEN RISK ESTIMATES..................... 7-88
7.3.1 Introduction....................................... 7-88
7.3.2 Unit Risk for Air.................................. 7-90
7.3.2.1 Methodology for Quantitative Risk Estimates.. 7-90
7.3.2.2 Risk Estimates from Epidemiologic Studies.... 7-94
7.3.2.2.1 The Lee-Feldstein (1983) Study.... 7-95
7.3.2.2.2 The Higgins et al. (1982) Study... 7-104
7.3.2.2.3 The Brown and Chu Estimates from the Anaconda Data............... 7-110
7.3.2.2.4 The Enterline and Marsh (1982) Study........................... 7-118
7.3.2.2.5 The Ott et al. (1974) Study....... 7-128
7.3.2.3 Discussion.................................. 7-132
7.3.3 Unit Risk for Water................................ 7-135
7.3.4 Relative Potency................................... 7-141
7.3.5 Summary and Conclusions of the Carcinogenicity of Arsenic.. 7-145
7.3.5.1 Qualitative Summary......................... 7-145
7.3.5.2 Quantitative Summary........................ 7-148
7.3.5.3 Conclusions................................. 7-148

TABLE OF CONTENTS
(continued)

Page

8. ARSENIC AS AN ESSENTIAL ELEMENT.. 8-1

9. HUMAN HEALTH RISK ASSESSMENT FOR ARSENIC.............................. 9-1
  9.1 AGGREGATE EXPOSURE LEVELS TO ARSENIC IN THE U.S. POPULATION.. 9-1
  9.2 SIGNIFICANT HUMAN HEALTH EFFECTS ASSOCIATED WITH AMBIENT EXPOSURES... 9-4
    9.2.1 Acute Exposure Effects............................... 9-4
    9.2.2 Chronic Exposure Effects............................. 9-5
  9.3 DOSE-EFFECT/DOSE-RESPONSE RELATIONSHIPS.................... 9-6
    9.3.1 General Considerations............................... 9-6
    9.3.2 Effects and Dose-Response Relationships.............. 9-8
      9.3.2.1 Respiratory Cancer............................ 9-8
      9.3.2.2 Skin Cancer................................... 9-9
      9.3.2.3 Non-cancerous Skin Lesions.................... 9-9
      9.3.2.4 Peripheral Neuropathological Effects and Cardiovascular Changes..................... 9-10
  9.4 POPULATIONS AT SPECIAL RISK TO ARSENIC EXPOSURE............ 9-10

10. REFERENCES.. 10-1

LIST OF TABLES

Table | Page

3-1 Physical Properties of Arsenic and Arsenic Compounds 3-2
3-2 Cumulative Frequency Distribution of NASN Individual 24-Hour Ambient Air Arsenic Levels 3-10
3-3 Primary Copper Smelters in the United States 3-13
3-4 Levels of Arsenic by Food Class in Adult Food Composites from 20 U.S. Cities 3-17
3-5 Levels of Arsenic by Food Class in Infant and Toddler Food Composites from 10 U.S. Cities 3-18
3-6 A Comparison of Arsenic Levels in Arsenic-Treated and Uncontaminated Soils in North America 3-21

5-1 Prevalence of Blackfoot Disease (per 1000) by Age and Arsenic Exposure 5-9

6-1 Summary of Mutagenicity of Arsenic: Gene Mutations 6-2
6-2 Summary of Mutagenicity of Arsenic: Chromosomal Aberrations 6-9
6-3 Summary of Mutagenicity of Arsenic: Chromosomal Aberrations and SCE in Cells of Exposed Humans 6-17
6-4 Mutagenicity of Arsenic: Assays Indicative of DNA Damage or Repair. 6-24
6-5 Summary of Mutagenicity of Arsenic: Interactions With Mutagens 6-33

7-1 Summary of Case Reports and Epidemiologic Studies of Cancer or Precancerous Lesions in Persons Exposed to Arsenic 7-2
7-2 Observed and Expected Deaths Due to Respiratory Malignancies, By Exposure Category 7-15
7-3 Observed and Expected Deaths for Selected Causes in Retrospective Cohort Analysis (1940-1973) 7-17
7-4 Observed and Expected Deaths and Standardized Mortality Ratios for Selected Causes of Death of 527 Males of Cohort Under Study 7-26
7-5 Observed and Expected Respiratory Cancer Deaths and Standardized Mortality Ratios by Arsenic Exposure Index 7-27
7-6 Observed and Expected Respiratory Cancer Deaths and Standardized Mortality Ratios by Intensity and Duration of Exposure to Arsenic... 7-27
7-7 Respiratory Cancer Deaths and SMRs By Cumulative Arsenic Exposure Lagged 0 and 10 years, Tacoma Smelter Workers 7-29
7-8 Respiratory Cancer Deaths and SMRs by Duration of Exposure and Latency, Tacoma Smelter Workers 7-30
7-9 Respiratory Cancer Deaths and SMRs by Duration and Intensity of Exposures, Tacoma Smelter Workers 7-32
7-10 1965 Smelter Survey Atmospheric Arsenic Concentrations 7-36
7-11 Observed and Expected Deaths from Respiratory Cancer, with Standardized Mortality Ratios (SMR), by Cohort and Degree of Arsenic Exposure, 1938-63 7-37
7-12 Mortality for All Causes and Respiratory Cancer from 1938 to 1978 by Time-Weighted Average (TWA) Arsenic Exposure as of Entrance into Cohort 7-44
7-13 Mortality for All Causes and Respiratory Cancer by Ceiling Arsenic Exposure as of Entrance into Cohort 7-44
7-14 Respiratory Cancer Mortality by Method of Analysis and TWA Arsenic Category 7-45

LIST OF TABLES (continued)

Table | Page
7-15 Respiratory Cancer Mortality by Method of Analysis and Ceiling Arsenic Category 7-46
7-16 Prevalence of Skin Cancer (per 1000) by Age and Arsenic Exposure (ppm) 7-54
7-17 Results of Total Arsenic Analysis and Arsenite and Arsenate Determination in the Yenshei Water Samples 7-57
7-18 Lane County Water Arsenic Levels 1974-1978 7-63
7-19 Age Specific Death Rates for Utah and Three Millard County Communities 7-67
7-20 Summary Table of Experimental Studies of Arsenic Carcinogenesis 7-78
7-21 Observed and Expected Deaths Due to Selected Causes, With Standardized Mortality Ratics (SMRs) Among Smelter Workers, 1938-1977 7-96
7-22 Description of Length of Employment Groups, With Numbers of Smelter Workers, Numbers of Deaths, Person-Years at Risk, and Duration of Smelter Employment (Based on Total Work Experience Through September 30, 1977) 7-97
7-23 Observed and Expected Deaths from Respiratory Cancer, With Person-Years of Follow-Up, By Cohort and Degree of Arsenic Exposure 7-98
7-24 Dose-Response Data from Lee-Feldstein (1983) Used for Risk Assessment 7-100
7-25 Summary of Quantitative Risk Analyses 7-103
7-26 Respiratory Cancer Mortality 1938-1978 from Cumulative Exposure to Arsenic for 1800 Men Working at the Anaconda Copper Smelter 7-106
7-27 Observed and Expected Lung Cancer Deaths and Person-Years By Level of Exposure, Duration of Employment and Age at Initial Employment 7-113
7-28 Arsenic Exposures: 1965 Smelter Survey Atmospheric Arsenic Concentrations 7-115
7-29 Observed and Expected Number of Respiratory Cancer Deaths for Each Cell in the Low-Exposure Group of Table 7-27 7-118
7-30 Cells from Table 7-29 Combined Within Rows to Obtain Cells With Three or More Expected Respiratory Cancer Deaths 7-119
7-31 Cells from Table 7-29 Combined Within Columns to Obtain Cells With Three or More Expected Respiratory Cancer Deaths 7-119
7-32 Observed Deaths and SMRs for 2802 Smelter Workers Who Worked a Year or More 1940-64, Followed Through 1976, By Cause of Death 7-120
7-33 Data From Table 8 of Enterline and Marsh (1982), With Person-Years of Observation Added 7-122
7-34 Data From Table 4 of Ott et al. (1974) 7-129
7-35 Combined Unit Risk Estimates for Absolute-Risk Linear Models 7-134
7-36 Age-Exposure-Specific Prevalence Rates for Skin Cancer 7-137
7-37 Data Utilized to Obtain Predictor Equation and Figure 7-11 7-138
7-38 Relative Carcinogenic Potencies Among 52 Chemicals Evaluated by the Carcinogen Assessment Group as Suspect Human Carcinogens 7-143

9-1 Routes of Daily Human Arsenic Intake 9-2

LIST OF FIGURES

Figure		Page
3-1	The Generalized Geochemical Cycle for Arsenic	3-5
3-2	Biological Cycling of Arsenic	3-7
3-3	NASN Annual Average Arsenic Concentrations	3-11
4-1	Arsenic Retention in Rat Lungs Following Intratracheal Instillation of a Single Dose	4-4
4-2	Lung Concentrations of Arsenic in Hamsters Given Weekly Intratracheal Instillations of Arsenic Trioxide, Arsenic Trisulfide or Calcium Arsenate	4-7
6-1	The Induction of Chromosome Aberrations in Cultured Human Peripheral Lymphocytes by Six Arsenic Compounds	6-14
7-1	Comparison of Census Tracts Experiencing Exposure to the Fisher Formation and Exhibiting High Skin Cancer Occurrence	7-64
7-2	Relative Risks and 90% Confidence Limits for Data of Lee-Feldstein (1983)	7-101
7-3	Absolute Risks and 90% Confidence Limits for Data of Lee-Feldstein (1983)	7-102
7-4	Relative Risks and 90% Confidence Limits for Data of Higgins (1982)	7-108
7-5	Absolute Risks and 90% Confidence Limits for Data of Higgins (1982)	7-109
7-6	Relative Risks and 90% Confidence Limits for Zero-Lag Data of Enterline and Marsh (1982)	7-124
7-7	Relative Risks and 90% Confidence Limits for 10-Year Lag Data of Enterline and Marsh (1982)	7-125
7-8	Absolute Risks and 90% Confidence Limits for Zero-Lag Data of Enterline and Marsh (1982)	7-126
7-9	Absolute Risks and 90% Confidence Limits for 10-Year Lag Data of Enterline and Marsh (1982)	7-127
7-10	Relative Risks and 90% Confidence Limits for Data of Ott et al. (1974) With Highest Exposure Group Omitted	7-131
7-11	Relationship Between Transformed Prevalence and Log ppm Arsenic in Water, Log Age	7-139
7-12	Histogram Representing the Frequency Distribution of the Potency Indices of 52 Suspect Carcinogens Evaluated by the Carcinogen Assessment Group	7-142

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1. INTRODUCTION

This report evaluates health effects associated with arsenic exposure, with particular emphasis placed on health effects thought to be of most concern in regard to exposure to inorganic arsenic of the general U.S. population. Organic arsenic compounds are considered only insofar as certain forms of the element arise via transformation of arsenic in man and other species, or arise by environmental transformation.

This report is organized into chapters which provide a cohesive discussion of all aspects of inorganic arsenic and delineate a logical linking of this information to human health risk. The chapters include: an executive summary (Chapter 2) of the information contained within the text of later chapters; background information on the chemical and environmental aspects of arsenic, including levels of arsenic in media with which U.S. population groups come into contact (Chapter 3); arsenic metabolism, where factors of absorption, biotransformation, tissue distribution, and excretion of inorganic arsenic are discussed with reference to the toxicity of the element (Chapter 4); arsenic toxicology, discussing the various acute, subacute, and chronic health effects of inorganic arsenic in man and animals (Chapter 5); arsenic mutagenesis, discussing the ability of inorganic arsenic to cause gene mutations, chromosomal aberrations, sister chromatid exchanges and inhibition of DNA synthesis (Chapter 6); arsenic carcinogenesis, including discussion of selected dose-effect and dose-response relationships (Chapter 7); arsenic as an essential element, which deals with the current status of inorganic arsenic as a required nutrient in at least some species of animals (Chapter 8); and a human health risk assessment for arsenic, where key information from the preceding chapters is placed in an interpretive and quantitative perspective highlighting those health effects of most concern for U.S. populations (Chapter 9).

This report is not intended to be an exhaustive review of all the arsenic literature, but is focused upon those data thought to be most useful and relevant for human health risk assessment purposes. Particular emphasis is placed on delineation of health effects and risks associated with exposure to airborne arsenic, in view of the most immediate use intended for the present report, i.e., to serve as a basis for decision making regarding the regulation of inorganic arsenic as a hazardous air pollutant under pertinent sections of the Clean Air Act, as amended in 1977. Health effects associated with the ingestion of arsenic or with exposure via other routes are also discussed, providing a basis for possible use for multimedia risk assessment purposes, as well. The background information provided at the outset on sources, emissions, and ambient concentrations of arsenic in various media is presented in order to provide a general perspective against which to view the health effects evaluations contained in later chapters of the document. More detailed exposure assessments, taking into account even more recent, up-to-date emission and ambient concentration data have been prepared separately for use in subsequent EPA regulatory decision making regarding arsenic.

The Agency recognizes that the regulatory decision-making process is a continuous one; therefore, should new information become available in the future that would warrent a reevaluation of the information contained herein, the Agency will undertake such an evaluation as part of its mandate to protect the health of the general U.S. population.

2. SUMMARY AND CONCLUSIONS

As a toxic agent, inorganic arsenic possesses several properties that are not shared with many environmental pollutants. The element exists in various chemical states, e.g., tri- and pentavalent inorganic arsenic and methylated organic arsenic, with each having differing toxicological potential. In man, experimental animals, and other organisms, arsenic undergoes a variety of transformations, the full significance and mechanisms of which are, as yet, not well understood. Furthermore, there appears to be a nutritional requirement for low levels of arsenic in certain experimental animals and this may also be the case for man. All of these factors complicate the analyses of the toxicological effects and the risk for human health associated with environmental exposure to arsenic compounds. The following chapter summarizes these factors as they are presented in depth in the ensuing document text.

2.1 BACKGROUND INFORMATION

2.1.1 Chemical/Physical Aspects of Arsenic

It is the various compounds of arsenic which have been of most importance in the extensive history of the toxicology of the element, the zero-valent metallic form being of minor toxicological interest.

Arsenic is encountered as a component of sulfidic ores of metals such as copper, cobalt and nickel. The smelting of these ores is associated with arsenic release into the environment. Arsenic trioxide, As_2O_3, a toxicologically significant form, is a smelter product arising from air roasting of these sulfidic ores.

Arsenic trioxide, white arsenic, is only sparingly soluble in water and other solvents which do not promote chemical transformation. The compound dissolves in acidic or alkaline aqueous media to yield either the free acid or salts, with these being soluble in a number of solvents. The oxide readily sublimes (135°C), and this factor is of importance in considering analytical methods for measuring levels of the compound.

The pentavalent arsenic pentoxide, As_2O_5, may be prepared by nitric acid oxidation of the trioxide or the element itself. This form has high solubility in water (63 g/100 g water), forming the strongly oxidizing arsenic acid, H_3AsO_4 ($E^o = 0.56V$).

Stability of the valency forms of arsenic in solution is dependent on the nature of the medium. Oxygenated media and higher pH favor the pentavalent form, while reducing and/or acidic media favor the trivalent state.

The acids of both valency forms of arsenic readily form alkali and alkaline metal salts, with the former being much more soluble than the latter. Organic ester derivatives of arsenic are quite labile to hydrolysis and this chemical behavior has biochemical/toxicological implications in the postulated role of arsenate ion in interfering with phosphorylation reactions.

Arsine (arsenic trihydride, AsH_3) is the most poisonous of the arsenicals, being a strong hemolytic agent, and it can be formed under certain restricted conditions, i.e., reduction of the oxy compounds in the presence of a strong hydrogen source.

Monomethyl and dimethyl arsenic arise by both environmental and in vivo transformation processes.

In high-temperature processes, arsenic is released as a vapor which is then adsorbed or condensed onto small particles. Such adherence to particles of 1-2 μm or less may result in enhanced health risk from the agent since particles in this size range are inhaled and deposited in the deepest part of the respiratory tract.

Arsenic compounds tend to form insoluble complexes with soils and sediments. In the case of soils, the interaction occurs with amorphous aluminum or iron oxides.

2.1.2 The Environmental Cycling of Arsenic

Primary smelting, biocide use and glass manufacturing constitute some of the major inputs of arsenic into the various environmental compartments. Of an estimated total release of approximately 10,000 short tons annually in the U.S., smelter activity accounts for about 50 percent, biocide (pesticide, fungicide, herbicide) use contributes 32 percent, and glass production contributes about 7.0 percent, with the remaining amount being released from various other sources.

The atmosphere is a major conduit for arsenic emitted from anthropogenic sources to the other environmental compartments via wet and dry precipitation processes. Dry and wet fall onto soils may be followed by movement through soils either into groundwater or surface water. Passage of arsenic into surface waters may be followed by transfer to sediments. Such cycling is made complex by chemical and biological transformations which have been reported as occurring in the various environmental compartments.

Trivalent arsenic in the atmosphere or in aerated surface waters can undergo oxidation to the pentavalent state, while pentavalent arsenic in media which are below pH 7.0 and contain oxidizable material can react to form the trivalent form via reduction.

Biological transformations of arsenic have been documented as occurring via both sedimentary bacteria and suspended marine algae. Several different hypotheses have been advanced to explain the biological cycling of arsenic.

Reduction and methylation of inorganic arsenic occur only to a limited extent in soils, one report noting a conversion of only 1-2 percent over a period of months.

With reference to the relative amounts of the annual environmental burden of arsenic, it has been calculated for 1974 that land is the major sink for arsenic, approximately 90 percent, with the atmosphere accounting for 7-8 percent and the smallest quantity appearing in waters.

2.1.3 Levels of Arsenic in Various Media

Available data on levels of arsenic in various media with which man interacts are generally presented as total arsenic, with limited information available for identifying specific chemical forms of arsenic.

2.1.3.1 Levels of Arsenic in Ambient Air--Based on the comprehensive data for U.S. air levels of arsenic obtained by the U.S. EPA's National Air Sampling Network, air levels of arsenic in the U.S. generally do not exceed 0.1 μg/m^3.

Generally, airborne arsenic is adhered to particulate matter. Although the immediate areas around smelters may contain some arsenic in the vapor form, data are available to indicate rapid adherence to particulate matter when sampling 2-3 km from these emission sites.

The specific chemical form(s) of airborne arsenic is still unclear. Generally, in most urban/suburban areas, arsenic is mainly in the form of a mixture of inorganic arsenic in the tri- and pentavalent states. Only in areas where methylated arsenic is used agriculturally or where biotic transformation can occur has methylated arsenic been found in air samples.

2.1.3.2 Levels of Arsenic in Drinking Water--The National Interim Primary Drinking Water Regulations, promulgated under the Safe Drinking Water Act, set the Maximum Contaminant Level (MCL) for arsenic in U.S. public water supplies at 50 μg/l. In general, arsenic is not found in drinking water at levels exceeding this MCL. Well waters in the western U.S. and Alaska, however, may have much higher levels owing to geochemical enrichment. In Lane County, Oregon, recent analyses report levels up to 2.2 ppm (2.2 mg/liter), while the highest figure in Alaska was 10 ppm (10 mg/liter), representing both natural and mining residue contributions.

It is reasonable to assume that the chief chemical form of arsenic in most public water supplies would be the pentavalent inorganic form, owing to both aeration and chlorination. Similarly, well waters in Alaska and the western U.S. are reported to mainly contain pentavalent inorganic arsenic.

2.1.3.3 Arsenic in Food--The most recent data base for the arsenic content of foods is the 1975-1976 survey carried out by the U.S. Food and Drug Administration. Shellfish and other marine foods have the highest levels on a food category basis. Overall, the total dietary intake of arsenic in 1975-1976 was approximately 50 μg (elemental arsenic), representing an increase from the preceding years. Whether this increase represents a trend or merely reflects analytical variation in sampling from year to year is still to be determined.

The chemical forms of arsenic in foods are varied and complex. Crustaceans and other marine life store arsenic in complex organoarsenical forms which, based on recent reports, are assimilated by man and generally excreted intact. Toxicologically, these forms are comparatively inert.

2.1.3.4 Arsenic in Soils--Background soil arsenic levels range from less than 1 ppm to over 40 ppm, the latter reflecting agricultural practices as well as air fallout. Soil arsenic is usually bound to clay surfaces, and its mobility is a function of soil pH, phosphate levels, iron and aluminum content, and soil type. The mobile fraction, usually in the pentavalent form, is of concern in terms of movement to plants and water. Little reductive methylation occurs in most soils.

2.1.3.5 Other Sources of Arsenic--Limited data on arsenic content of tobacco suggest that more recent values range from around 1.5 ppm or less, while in the past (1945) values up to 40 ppm were measured. This decrease reflects reduced use of arsenical biocides in tobacco production.

2.2 ARSENIC METABOLISM

2.2.1 Routes of Absorption

Major routes of absorption of arsenic in the general population are inhalation and ingestion. The ingestion of arsenic may be either through the direct intake of food and water or through secondary intake via inhalation and swallowing. Arsenic uptake through the skin appears to be a minor route of exposure. Factors affecting the extent of absorption include chemical forms, particle size, and solubility.

2.2.1.1 Respiratory Absorption--Limited data from human subjects suggest that about 40 percent of most forms of inhaled arsenic is deposited in the lungs, all of which is eventually absorbed, yielding a net absorption of approximately 30 percent or greater of the inhaled amount.

Several studies of smelter workers also confirm that significant absorption of inhaled arsenic occurs, as judged by rapidly rising urine arsenic levels when exposure first occurs. Furthermore, the levels excreted are correlated with workplace air levels.

Animal data also indicate respiratory tract absorption when exposure occurs by inhalation of aerosols or by intratracheal instillation.

2.2.1.2 Gastrointestinal Absorption--Based on a number of reports using human subjects, soluble inorganic arsenic is almost totally absorbed from the gastrointestinal tract. Similar data have been obtained for such experimental animal species as the rat, pig, and monkey.

Less soluble forms of arsenic, such as arsenic trioxide in suspension, lead to considerably lower absorption while insoluble arsenic triselenide passes through the human GI tract with negligible absorption.

2.2.1.3 Transplacental Transfer--Transplacental transfer of arsenic in man appears to occur, based on autopsy data and on one report showing that newborn cord blood levels approximate those of the mothers. Measurable levels of arsenic in fetal tissue have been determined by the fourth month of gestation, increasing to month seven.

In animals, inorganic arsenic appears to rapidly cross the placental barrier where it is distributed in embryonic tissue.

2.2.2 Biotransformation of Inorganic Arsenic In Vivo

An understanding of inorganic arsenic metabolism in man and other species is complicated by recently revealed biotransformation processes, discovered because of the development of analytical techniques which permit the chemical speciation of arsenic into its various forms. These processes not only relate to pharmacokinetic parameters such as tissue distribution and excretion, but also figure in the toxicology of the element. The two processes of significance for consideration here are the methylation of inorganic arsenic and oxidation-reduction interconversion of inorganic arsenic.

An extensive recent literature documents the *in vivo* methylation of inorganic arsenic to monomethyl and dimethyl arsenic (the latter being the major methylated metabolite) in every mammalian system studied to date, including man.

While the quantitative features of this phenomenon may vary among species, one can generally state that: 1) dimethyl arsenic is the major transformation product in both humans and animals, with the amount of monomethyl arsenic, when formed, being greater in humans; 2) methylation is assumed to be a route of detoxification, the methylated forms being not only much less toxic but also more rapidly excreted; 3) the methylating capacity of a given system can persist over a range of inorganic arsenic exposure but at some point can be overwhelmed; and 4) retrospective assessment of early data on arsenic metabolism must be reviewed in light of the current knowledge about biomethylation. In man, dimethyl arsenic represents approximately 75 percent of total arsenic excretion, with monomethyl arsenic being excreted in lesser amounts.

The demonstration of interconversion of the two valency forms of inorganic arsenic claimed in earlier literature must be considered in light of the biomethylation phenomenon, and only more recent studies addressing this problem using chemical speciation techniques can be considered reliable. Invariably, the giving of inorganic penta- or trivalent arsenic to experimental animals or human volunteers leads to predominantly methylated forms, with any inorganic arsenic being present in small amounts. These observations and supporting data showing reduction of pentavalent arsenic _in vivo_ are consistent with a reductive biomethylation mechanism, including initial reduction of pentavalent to trivalent arsenic prior to methylation. A recent study using speciation methods and human subjects also establishes the _in vivo_ reduction of pentavalent to trivalent arsenic in humans.

Even with sophisticated analytical methods permitting chemical speciation in biological media, the lability of arsenic in dosing solution to oxidation-reduction requires careful assessment of the actual form being administered.

2.2.3 Distribution of Arsenic in Man and Animals

Blood is the main vehicle for transport of arsenicals following absorption, and from which arsenic is cleared relatively rapidly to tissues in all species except the rat. In the rat, arsenic-erythrocyte interaction tends to preclude rapid movement to tissues, with a biological half-time of up to 90 days, versus a corresponding time of several days in all other species. Arsenic movement from blood appears to conform to a three-compartment model which must reflect in part the biomethylation of inorganic arsenic noted above.

Exposure of various experimental animals to either tri- or pentavalent inorganic arsenic leads to initial accumulation of the element in liver, kidney, spleen, aorta, and skin. In most species, arsenic clearance from soft tissue is relatively rapid except for the skin, where the high sulfhydryl group content probably promotes tight arsenical binding.

In man, tissue partitioning data are mainly available from autopsy data. Heart, kidney, liver and lung have highest levels on a concentration basis, but skin and excretory/storage organs such as nails and hair have the highest absolute amounts. Brain tissue has levels only slightly below those of other soft tissues.

Recent data on valency and exposure level effects on tissue distribution of arsenic indicate that levels of arsenic in kidney, liver, bile, brain, skeleton, skin, and blood are 2- to 25-fold higher for the trivalent form than for the pentavalent state, and are greatly increased at higher dosing. The difference is held to be due to the relative methylating capacity of either form as well as the level of exposure.

2.2.4 Arsenic Accumulation

The long-held view of arsenic as an element which accumulates in the body was mainly based on the behavior of arsenic in the rat, an anomalous model which does not reflect the case with other species. For other species, including man, long-term accumulation does not generally occur in physiologically active compartments in the body.

In man, the only evidence for tissue accumulation is autopsy data from retired workers with a history of metal smelter exposure in which lung arsenic levels were 8-fold higher than in a control population. This suggests the existence of a very insoluble form of arsenic in smelter ambient air.

2.2.5 Arsenic Excretion

Renal clearance appears to be the major route of excretion of absorbed arsenic in man and animals. Biliary transport of the element leads to enteric reabsorption, with little carriage in feces.

In man, inorganic arsenic is excreted rather rapidly, and in several studies in which continuous exposure resulted in the acquisition of steady state, around 60 percent of a given dose was excreted within one day.

The pattern of renal excretion reflects the *in vivo* biotransformation capacity for inorganic arsenic, one study noting that trivalent arsenic was excreted more slowly than an equivalent dose of the pentavalent form, and higher doses of both forms were cleared relatively more slowly than lower doses.

2.3 ARSENIC TOXICOLOGY

2.3.1 Acute Toxicity

Acute symptoms of arsenic poisoning are similar in both man and experimental animals. With oral exposure, the acute symptoms include severe gastrointestinal damage resulting in vomiting and diarrhea and general vascular collapse leading to shock, coma, and death. Other acute effects are muscular cramps, facial edema, and cardiovascular reactions. Airborne exposure at high levels also results in severe irritation of nasal mucosa, larynx, and bronchi. Sequelae of acute contact with inorganic arsenic include peripheral nervous system disturbances with slow recovery and reversible effects on the hematopoietic system.

Levels of exposure associated with acute arsenic toxicity vary with the valency form of the element, the trivalent state being approximately 4-fold more toxic than pentavalent arsenic. Oral LD_{50} values for trivalent arsenic vary from 15 to 293 mg/kg b.w. in rats and from 10-150 mg/kg in other test species.

While a number of outbreaks of acute arsenic poisoning have been described, few data exist on actual doses and type of arsenical involved. One report has estimated the human lethal dose for arsenic trioxide to be anywhere from 70 to 180 mg.

2.3.2 Chronic Toxicity

Two categories of chronic arsenic toxicity can be discerned from the available literature: 1) the mutagenicity/carcinogenicity of arsenic; and 2) various non-carcinogenic chronic effects.

2.3.2.1 Mutagenesis of Inorganic Arsenic--Various inorganic compounds of arsenic have been tested for mutagenicity in a variety of test systems ranging in complexity from bacteria to peripheral lymphocytes of exposed human beings. Although many of the data are contradictory, the weight of evidence supports the following conclusions:

1) Arsenic is either inactive or extremely weak for the induction of gene mutations *in vitro*.
2) Arsenic is clastogenic and induces sister chromatid exchanges (SCE) in a variety of cell types, including human cells *in vitro*; trivalent arsenic is approximately an order of magnitude more potent than pentavalent arsenic.
3) Arsenic does not appear to induce chromosome aberrations *in vivo* in experimental animals.
4) Several studies suggest that human beings exposed to arsenic demonstrate higher frequencies of SCE and chromosomal aberrations in peripheral lymphocytes; however, the quality of these studies is generally poor.

5) Arsenic may affect DNA by the inhibition of DNA repair processes or by its occasional substitution for phosphorus in the DNA backbone.

From these conclusions, it can be surmised that arsenic may have the potential to cause chromosomal changes in human beings. Although additional information is needed, it appears that the mutagenic potency of arsenic is weak compared to other known metal mutagens.

2.3.2.2 Carcinogenesis of Inorganic Arsenic--The current status of inorganic arsenic as a human and experimental animal carcinogen has been extensively and critically reviewed by public agencies such as the National Institutes of Occupational Safety and Health, scientific bodies such as the National Academy of Science and the International Agency for Research on Cancer, and in a number of individual assessments.

At present, the collective evidence for an etiological role of inorganic arsenic in human cancers is strongest for cancers of the skin and lung.

In man, chronic oral exposure to arsenic induces a sequence of changes in skin epithelium, proceeding from hyperpigmentation to hyperkeratosis characterized as keratin proliferation of a verrucose nature, and leading in some cases to late onset skin cancers. The skin cancers can be histopathologically characterized as either squamous carcinomas in the keratotic areas or basal cell carcinomas.

Arsenic-associated skin cancers, regardless of type of exposure, differ from those having an ultraviolet light etiology in that they occur on unexposed areas such as the palms and soles and occur as multiple lesions. The latency period for such lesions has been reported to range from 6 to 50 years for skin cancer induced by the administration of arsenical medicinals. The most reliable study of skin cancer associated with arsenic-contaminated drinking water found the minimum human latency period to be 24 years.

Lung cancers associated with occupational exposure to arsenic appear to be mainly of the poorly differentiated and small-cell undifferentiated epidermoid carcinoma type, although well-differentiated epidermoid carcinoma and acinar-type adenocarcinoma have also been noted. The latency period for such lung cancer due to occupational arsenic exposure at smelters has been reported to range from 13 to 50 years.

While other visceral cancers such as hepatic angiosarcomas have also been claimed to be associated with arsenic exposure, the data base for such association is less conclusive than for cancers of the skin and lung.

2.3.2.2.1 Human Epidemiology of Arsenic Carcinogenesis. Cancer- and possible precancerous lesion-producing inorganic arsenic exposures have been demonstrated in both occupational populations, such as copper smelters, pesticide manufacturers and argricultural workers, and in non-occupational populations using arsenical drugs or consuming arsenic-contaminated drinking water and/or food.

Cancer or possible precancerous skin lesions associated with arsenic exposure include lung cancer, skin cancer, peripheral vasculopathy, hyperkeratosis, hyperpigmentation, and depigmentation. Of these, cancers of the lung and skin have received the most attention. Furthermore, epidemiologic reports of cancer or possible precancerous lesions related to arsenic exposures have a wide geographical distribution, with studies reported in the United States, Europe, Asia and South America.

Lung Cancer

An excess mortality in respiratory cancer has been noted among smelter workers and among workers engaged in the production and use of arsenical pesticides. In a number of these studies, the levels of exposure are uncertain and there is simultaneous exposure to agents such as other metals and sulfur dioxide. Furthermore, some of these studies did not take into account the effects of cigarette smoking.

A proportionate mortality study of an English factory which manufactured arsenite as a sheep dip powder found that workers at the facility had a proportion of lung cancer deaths twice that for other workers in the geographic area. Of the total factory group, the chemical workers, who were the most closely associated with the arsenite production, accounted for all of the lung cancer deaths and had a higher proportion of deaths from all cancers than the total factory population. Air exposure information was limited, levels up to 4 mg As/m^3 having been reported only for 1945-1946. The outcome of the earlier English study has been supported by studies of two facilities in the United States. At one facility manufacturing lead-, calcium-, and magnesium arsenate and copper acetoarsenite over the period 1919-1956, the ratio of observed-to-expected lung cancer deaths among workers was evaluated on the basis of exposure level, giving ratios ranging up to 7:1 in the highest exposure category.

The second U.S. facility, in Baltimore, Maryland, has been the subject of two occupational studies. At this operation, production of lead-, calcium-, and sodium arsenate was started in the early 1950s. Air levels of arsenic were not available. In the most recent report--a follow-up of workers employed from 1946 to 1974--statistically significant increases in lung cancer deaths were found in "predominantly arsenical production workers." Information on smoking history was not obtained.

Occupational exposure to arsenic also occurs in smelters. Several reports have centered on workers at a copper smelter in the state of Washington. In the most recent publication concerning cancer deaths at this facility, an increase in overall mortality, a significant increase in cancer deaths, and a highly significant increase in deaths from lung cancer were noted. Although cigarette smoking data were not available, a clear lung cancer dose response by arsenic exposure was found, suggesting that smoking probably did not confound the lung cancer response believed to be due to arsenic.

Other U.S. studies have been done at smelter sites in Utah and Montana. In the Utah study, smelter workers showed a 3-fold increase in the lung cancer death rate compared to the general population of the state. Exposures to sulfur dioxide and copper, as well as arsenic, were found to be significantly higher for the lung cancer cases. Differences in smoking habits could not explain the excess lung cancer mortality. Smelter workers at the Montana smelter were found to follow a lung cancer dose response by arsenic exposure. Differences in smoking habits could not explain the differences in lung cancer mortality. Little association, if any, was found between sulfur dioxide exposure and lung cancer mortality.

Some of the evidence from the U.S. smelter studies suggests that arsenic may be acting as either a late-stage carcinogen in the multistage theory of carcinogenesis or as a promoter in the carcinogenesis process. Either hypothesis requires further study, however.

Excess lung cancer mortality has also been reported for workers in foreign smelters. Several reports have concerned the smelter facility in Sweden. In this operation, it was found that arsenic exposure was associated with increases in lung cancer deaths, the ratio being 4.6 for a work history of more than 17 years. Sulfur dioxide exposure was not found to be associated with lung cancer, nor could the results be explained by differences in smoking habits. A study of smelter workers in Japan has also reported a positive association between smelter employment and lung cancer. Workers in the highest exposure and longest follow-up category were reported to have a 25-fold increase in lung cancer mortality.

Several studies suggest that populations surrounding arsenic-emitting sources are at a greater risk of lung cancer. One study found that lung cancer in counties with smelters was significantly higher than in counties without smelters. Another study found an association between residence in an area surrounding an arsenic pesticide plant and lung cancer mortality even after controlling for employment at the pesticide plant. Two lung cancer mortality studies of populations in the vicinity of smelters are inconclusive with regard to lung cancer risk in the general population because lung cancer deaths of workers at the smelters were included in the analyses. It should be noted that none of these studies addressed the effects of population migration, however.

Skin Cancer and Precancerous Lesions

All of the occupational studies on arsenic have been mortality studies. Because skin cancer is rarely fatal, the occupational studies, in general, do not report excesses of skin cancer. The English study referred to in the preceding section, however, did note that cancer deaths among those factory workers with skin cancer was 13.6 percent compared to a figure of 1.3 percent in the reference population.

Most of the available information on the association of arsenic with skin cancer has involved non-occupational populations in geographical areas as diverse as Taiwan, Argentina, and northern Europe, and has involved arsenic in drinking water or medicinal preparations.

In Taiwan, exposure started in 1910-1920 with the availability of water from deep wells. In the most comprehensive study of this group, 37 villages with a population of 40,421 were surveyed in 1965 and the prevalence rate of skin cancer was categorized by arsenic exposure and age. The prevalence rate of skin cancer increased with both well water arsenic and age. The greatest prevalence rate was in the >60-years age group (192.0/1,000 subjects) exposed to well water with an arsenic content of >0.6 ppm. There was a similar ascending gradient for hyperkeratosis, an apparent precondition for the later onset of skin cancer.

In Cordoba, Argentina, arsenical waters have also been reported to be associated with increased rates of skin cancer among the population using this water. In the various reports on this area, it has been noted that over 81 percent of persons with "arsenical-type" skin epitheliomas presenting themselves at a particular dermatology clinic came from areas where high levels of arsenic were known to be present in the water supplies and where arsenicism among individuals was known to occur. Lack of information on the size of the populations in each geographic area limits the conclusions that can be drawn from these studies, however.

In a number of epidemiological surveys conducted in Antofagasta, Chile, the presence of a relatively high level of arsenic, 0.6 ppm, in the public drinking water from 1958 to 1970, was shown to have resulted in a high number of skin lesions including hyperkeratosis which may be a precondition for skin cancer. In one report of cases seen and diagnosed from 1968 to 1971 in Antofagasta, the author reported 457 patients with various cutaneous lesions including leukoderma, melanoderma, hyperkeratosis, and squamous-cell carcinoma; all had high arsenic content in the hair. The number of cases for each diagnosis was not specified.

An investigation of the population having contact with well waters containing elevated levels of arsenic in an area of Oregon did not yield any evidence for skin cancer associated with such exposure, nor did a similar study in Utah. Factors which may have contributed to the difference in results between these studies and the studies in Taiwan, where an association between skin cancer and arsenic in drinking water was found, include: (1) in addition to arsenic, ergotamine was found in the Taiwan drinking water, (2) the socioeconomic level of the Taiwanese who experienced skin cancer was rather low, which may have had ramifications in terms of diet, personal hygiene and medical care, and (3) racial differences between the Taiwanese and U.S. populations may have had an effect. However, the population sample sizes of the Oregon and Utah studies were too small to detect the risk from skin cancer that would have been predicted, on the basis of arsenic ingestion, from a linear model of the Taiwanese data.

The chronic ingestion of trivalent arsenic present in the medicinal preparation, Fowler's Solution, has been shown to be associated with the typical arsenic dermatopathology, including skin cancer. In one detailed study of patients with a history of Fowler's Solution use, 21 cases of skin cancer and 106 cases of hyperkeratoses were found among 262 subjects. Both the hyperkeratosis and skin cancer prevalence rates were found to increase as the total ingested amounts of the arsenical increased. The minimal latency period for hyperkeratosis was 2.5 years while the minimal latency period for skin cancer was 6 years, with an average of 14 years.

Internal neoplasms associated with arsenic exposure have been reported in subjects exposed to medicinal arsenic as well as in such occupational groups as vintners and smelter workers.

2.3.2.2.2 Experimental studies of arsenic carcinogenesis. Consistent demonstration of arsenic carcinogenicity in test animals, using different chemical forms, routes of exposure, and various experimental species, has not been observed. Part of the difficulty may lie in the selection of an appropriate animal model. The absence of skin cancers in the experimental animals used could be accounted for by their skin being poor models of human skin. Rat results must be considered suspect because the rat is anomalous in how it metabolizes arsenic.

Recent data indicate that tumorigenicity and possibly carcinogenicity can be demonstrated in animals if the retention of arsenic in the lung is increased. The additional observations that calcium arsenate, as reported from two laboratories, is only slowly cleared from the lung, strongly suggest that this agent may be carcinogenic.

2.3.2.3 Chronic Neurological Effects of Arsenic Exposure--Both peripheral and central nervous system effects have been documented in man and animals exposed to inorganic arsenic.

Peripheral nervous system effects have been noted in workers occupationally exposed to arsenic and in individuals accidentally exposed to food contaminated with arsenic. Some of these effects follow an insidious course, appearing months or years after onset of exposure. Effects are of both the sensory and motor types, sensory deficits manifesting themselves first. The resulting polyneuropathies tend to follow a slow course of recovery over months or years. More subtle neurological effects, such as neuromuscular disturbances and altered nerve conduction velocity, have also been reported by various investigators.

While there are documented cases of central nervous system (CNS) effects in children due to acute or subacute exposure, chronic arsenic intoxication as a factor in such abnormalities as hearing impairment has not been confirmed.

Few useful animal models exist for the central and peripheral nervous system effects seen in humans. One study has reported evidence of CNS functional deficits in rats exposed to arsenic trioxide aerosol, while a second report claims impaired behavioral responses in rats given arsenic trioxide via stomach intubation. Since rats handle arsenic differently than all other species, the significance of these data is not clear.

2.3.2.4 Cardiovascular Effects of Arsenic Exposure--Cardiovascular effects associated with chronic arsenic intake include Blackfoot disease seen in Taiwanese consuming well water having elevated arsenic. This is a peripheral vascular disease leading to gangrene of the extremities. Since ergotamine-like compounds were also present in the waters, the conclusive role for arsenic is not clear. Peripheral vascular changes have been documented among German vintners who were exposed occupationally to arsenic pesticides as well as arsenic in wine.

2.3.2.5 Other Systemic Effects of Arsenic--Non-cancerous respiratory effects of inorganic arsenic are mainly seen with occupational exposure and with arsenic trioxide. In one study of smelter workers, those handling refined arsenic showed nasal septum perforation and rhinopharyngolaryngitis, while workers in roaster, furnace, and convertor areas showed tracheobronchitis and pulmonary insufficiency.

Teratogenic effects of arsenic compounds at relatively high exposure levels have been demonstrated in a number of animal species, including hamsters, rats, and mice. Generally, such effects have been observed after parenteral administration of either arsenite or arsenate. Oral exposures of animals to these same arsenic compounds at lower doses, by contrast, have not been shown to produce any notable effects on reproduction and development. Methylated arsenic, while potentially teratogenic at high doses, is of lower potency than inorganic forms. There is little evidence that inorganic arsenic is a human teratogen.

Hepatic effects have been noted in a number of studies dealing with chronic intake of arsenic. These disturbances present as cirrhosis and portal hypertension. One complicating factor in occupational exposure assessment has been the effect of alcohol consumption.

Chronic exposure to arsenic via well water contaminated by arsenic, medicinals, or the workplace, produces a reversible anemia. Rats, mice, and cats fed either arsenite or arsenate show reduced hemoglobin production via disturbance of the ALA-synthetase and heme-synthetase steps in the heme biosynthetic pathway.

Renal effects seen with arsenite or arsenate exposure in either man or animals have been poorly characterized.

2.3.3 Factors Affecting Arsenic Toxicity

The most widely recognized and studied interactive behavior of arsenic is with selenium, the pair being antagonistic in effect in all animal species studied. Dietary arsenic supplementation is known to protect against selenium toxicity, while selenium protects against either tri- or pentavalent arsenic. Arsenite shows a protective effect for selenite toxicity in cell cultures, but the reverse does not appear to be the case.

Few data exist for interactive relationships between arsenic and other elements. One report notes that cadmium and arsenic given simultaneously by the oral route retarded weight gain in young adult rats to a greater extent than either element given alone.

2.4 ARSENIC AS AN ESSENTIAL ELEMENT

Inorganic arsenic appears to be an essential element in certain animal species--rats, goats, chicks, and minipigs--based on the observation of detrimental effects using diets deficient in the element.

In rats, arsenic-deficient diets in pregnant dams are associated with slow growth, enlarged spleens, erythrocyte dysfunction in post-weaning offspring, and greater perinatal mortality.

In studies using goats and minipigs, diets having less than 50 ppb As produced effects in the adult animals as well as in their offspring. Mortality of adult goats was increased as was the mortality of both kids and piglets. In chicks, arsenic deprivation influenced the effects of dietary arginine, manganese, and zinc, the fluctuations of which variously affected metabolic activity.

Remaining to be independently demonstrated are a physiological role for arsenic, the existence of any specific carrier in the body, or arsenic essentiality in man.

2.5 HUMAN HEALTH RISK ASSESSMENT FOR ARSENIC

This portion of the summary contains the data summarized earlier, presented within a perspective of the possible quantitative risk posed to the general population of the United States by arsenic exposure. Categories of consideration include: (1) levels of arsenic in media relevant to the U.S. population; (2) those effects relevant to the general population; (3) indicators of exposure, specifically "internal dose" measures; (4) dose-effect and dose-response relationships which can be determined from available information; and (5) the identification of groups within the general population who may be at increased risk for the health effects of arsenic.

2.5.1 Exposure Aspects of Arsenic

Arsenic exposure in the general population of the United States occurs via inhalation and ingestion of water and food.

Respiratory intake of arsenic on a daily basis is approximately 0.12 µg, of which 0.03 µg would be absorbed, assuming 30 percent absorption and based on a 1981 national average air value of 0.006 $\mu g/m^3$ of air and a daily ventilation rate of 20 m^3.

Since much of the U.S. drinking water supply is below the 10 µg/liter level, daily consumption of water at a rate of 2 liters would lead to an intake $\leq$ 20 µg.

Based on 1975-76 figures from the Food and Drug Administration, daily arsenic intake from food is approximately 50 μg (elemental arsenic) for adults. If one assumes at least 80 percent absorption of this daily intake, approximately 40 μg is the absorbed amount. It should be noted, however, that arsenic in certain foods is known to be in a chemically complex form that is relatively resistant to metabolism and toxicologically inert; consequently, it is rapidly excreted intact. Therefore, the amount of absorbed food arsenic considered to be toxicologically significant is relatively small compared to total arsenic intake.

Cigarette smoking contributes about 6 μg/pack of cigarettes in mainstream smoke, of which approximately 2.0 μg/pack would be absorbed.

Thus, the aggregate absorbed amount is approximately $\leq$ 60 μg for nonsmokers. However, the actual amount of toxicologically significant arsenic taken in daily would probably be closer to 20 μg or less.

2.5.2 Effect/Response Aspects of Arsenic

2.5.2.1 Relevant Health Effects--General population concerns are with effects arising from long-term exposure to moderate levels of arsenic.

One can rank health effects germane to the general population as follows:

1. Respiratory tract cancer.
2. Skin cancer.
3. Non-cancerous skin lesions.
4. Peripheral neuropathological effects.
5. Cardiovascular changes.

Cancer of the respiratory system is clearly associated with exposure to arsenic via inhalation. This association has been especially noted among workers engaged in the production and usage of pesticides and among smelter workers. The latency period for lung cancer due to occupational arsenic exposure has been reported to range from 13 to 50 years.

Cancer of the skin has been found as a dose-dependent effect in a Taiwanese population with lifetime exposure to well water arsenic as well as among people who were treated with large doses of arsenite for skin disorders. Arsenic-associated skin cancers occur on areas of the body generally unexposed to the ultraviolet light of the sun, such as the palms of hands and soles of feet. The latency period for skin cancer has been reported to range from 6 to 50 years for cancers induced via administration of arsenical medicinals. In the Taiwanese drinking water study, the minimum latency period was reported to be about 24 years.

The Carcinogen Assessment Group (CAG) of the U.S. Environmental Protection Agency (EPA) has concluded that there is sufficient evidence that inorganic compounds of arsenic are both lung and skin carcinogens in humans.

Hyperkeratosis and hyperpigmentation, sometimes with precancerous changes, have been a common finding in persons ingesting arsenic. These skin lesions, like the manifest cancer, develop on surfaces usually unexposed to sunlight.

Peripheral nervous system effects range from sensory-motor deficits with higher exposure to changes in electromyography and nerve conduction velocity at long-term, low levels of contact with arsenic.

Vascular effects, such as Blackfoot disease (peripheral vasculopathy) have been noted in a Taiwanese population having life-long arsenic exposure; however, these effects may have been confounded by the presence of ergotamine-like compounds. Peripheral vascular changes have also been found in German vintners exposed occupationally to arsenic pesticides as well as arsenic in wine.

2.5.2.2 Dose-Effect/Dose-Response Relationships--The general question of how to define and employ a dose factor in attempts at quantitative assessments of human health risk for any toxicant is highly dependent upon: 1) the available information on the body's ability to metabolize the agent, and 2) the assessment of the relative utility of various internal indices of exposure.

The time period over which a given total intake occurs is highly important. For example, intake of one gram of arsenic over a period of years would be quite different pathophysiologically from assimilating this amount at one time, the latter probably having a lethal outcome. This time-dependent behavior is related in part to the relative ability of the body to detoxify inorganic arsenic by methylation as a function of both dose and time.

In cases of acute and sub-acute exposure, indicators of internal exposures, such as blood or urine arsenic levels, are probably appropriate for assessing the intensity of exposure.

With chronic low-level exposure, however, the available data would indicate that the total amount assimilated is probably more important than an indicator concentration without knowledge of the total exposure period. An added problem is the background level of arsenic found in some indicators due to dietary habit. Therefore, in low-level chronic exposures, arsenic levels in blood or urine would only be moderately increased over background levels. Hair arsenic levels cannot be employed as reliable indicators of exposure because no methods exist for distinguishing external contamination levels from those accumulated via absorption and metabolic distribution.

Given the above limitations concerning the use of blood, urinary or hair arsenic concentrations as internal indices of cumulative, long-term low-level arsenic exposure, the dose-effect/dose-response relationships summarized below are done so mainly in terms of external arsenic exposure levels via either inhalation or ingestion.

From available data, the Carcinogen Assessment Group (CAG) has estimated carcinogenic unit risks for both air and water exposures to arsenic. The quantitative aspect of carcinogen risk assessment is included here because it may be of use in setting regulatory priorities, evaluating the adequacy of technology-based controls, and other aspects of the regulatory decision-making process. However, the imprecision of presently available technology for estimating cancer risks to humans at low levels of exposure should be recognized. At best, the linear extrapolation model used (see Section 7.3) provides a rough but plausible estimate of the upper limit of risk--that is, with this model it is not likely that the true risk would be much more than the estimated risk, but it could be considerably lower. The risk estimates presented below should not be regarded, therefore, as accurate representations of true cancer risks even when the exposures involved are accurately defined. The estimates presented may, however, be factored into regulatory decisions to the extent that the concept of upper-risk limits is found to be useful.

The air estimates were based on data obtained in five separate studies involving three independently exposed worker populations. Linear and quadratic response models in both the absolute and relative form were fitted to the worker data. It was found that for the models that fit the data at the $p = 0.01$ or better level, the corresponding unit risk estimates ranged from 1.05×10^{-4} to 1.36×10^{-2}. However, linear models were found to fit better than quadratic models and absolute models fit better than relative models. The CAG also felt that exposure to trivalent arsenic was more representative of low environmental exposure than pentavalent arsenic. Restricting their unit risk estimates to those obtained from linear absolute models where exposure was to trivalent arsenic gave a range of 1.25×10^{-3} to 7.6×10^{-3}. A weighted average of the five estimates in this range gave a composite estimate of 4.29×10^{-3}.

The unit risk estimates for water were based on an extensive drinking water study which was conducted in a rural area of Taiwan. An association between arsenic in well water and skin cancer was observed in the study population. Using the male population, who appeared to be more susceptible, the CAG estimated that the unit risk associated with drinking water contaminated with 1 µg/ℓ of arsenic was 4.3×10^{-4}.

To compare the air and water unit risks, the CAG converted the exposure units in both cases to mg/kg/day absorbed doses, which resulted in unit risk estimates of 50.1 and 15.0, respectively.

The potency of arsenic compared to other carcinogens was evaluated by noting that an arsenic potency of $2.25 \times 10^{+3}$ $(\text{mMol/kg/day})^{-1}$ lies in the first quartile of the 52 suspect carcinogens that have been evaluated by the CAG.

The U.S. EPA is presently examining information from studies on both patient and general populations which have been exposed to arsenic via medicinals or drinking water, respectively, in order to determine whether quantitative dose-response relationships can be established for non-cancerous skin lesions.

While the qualitative evidence for peripheral neurological effects and cardiovascular changes in arsenic-exposed populations is well established, the data are insufficient for determining quantitative dose-response relationships at the present time.

2.5.3 Populations at Special Risk to Health Effects of Arsenic

From a Japanese study, which reported on the poisoning of children exposed to arsenic in infant milk formula, young children may be considered at risk for acute exposure to arsenic. From the clinical reports published at the time of the mass poisoning, as well as those from follow-up studies, a number of signs of central nervous system involvement were noted at both the time of the episode and much later, with the follow-up studies showing behavioral problems, abnormal brain wave patterns, marked cognitive deficits, and severe hearing loss.

Because children consume more water per unit body weight than do adults, the daily intake of arsenic via drinking water per kilogram body weight would be greater in children. This might have implications regarding chronic exposure effects in children. However, it should be noted that serious health effects due to chronic exposure of arsenic in drinking water have not been found at a greater frequency in children than adults.

Individuals residing in the vicinity of certain arsenic-emitting sources, e.g., certain types of smelters, may be at risk for increased arsenic intake because of both direct exposure to arsenic in air and indirect exposure via arsenic secondarily deposited from air onto soil or other human exposure media. The relative contribution from such indirect exposures to increased risk would be difficult to define, however.

A less defined group at risk would be cigarette smokers due to some arsenic in tobacco, but it is not clear just what the quantitative increase in risk would be.

3. BACKGROUND INFORMATION

3.1 CHEMICAL/PHYSICOCHEMICAL ASPECTS

Compounds of arsenic in various chemical forms have most prominently figured in the extensive history of the toxicology of the element. The physical properties for several of these compounds are presented in Table 3-1. In contrast, the element in the metallic, zero-valent form is of minor toxicological interest.

Geochemically, arsenic is encountered as a component of sulfidic ores, occurring as the arsenides and diarsenides of metals such as nickel, cobalt, and copper, and is present in rocks and soils at trace levels. Smelting of commercially important metal ores, therefore, often has associated with it the release into the environment of significant quantities of certain arsenic compounds. For example, arsenic trioxide, As_2O_3, a major form of the element in terms of its toxicological history, is a smelter product arising from air roasting of metallic arsenides or arsenic-containing sulfides.

Arsenic trioxide, white arsenic, is only slightly soluble in water and other solvents which do not promote chemical transformation. Its solubility in solvents which mimic physiological media may not necessarily be the same as for simple solvents, e.g., gastric juice versus water. Arsenic trioxide sublimes, the process becoming pronounced at 135°C. This property appears to have been overlooked through the years in considering analytical methods for measuring levels of the compound. Dissolution of the trioxide in aqueous media leads to formation of arsenous acid, H_3AsO_3, while alkaline treatment leads to formation of the arsenite ion, $AsO(OH)^-{}_2$, with both the acid and the salt being freely soluble in a number of solvents.

TABLE 3-1. PHYSICAL PROPERTIES OF ARSENIC AND ARSENIC COMPOUNDS

Chemical name	Atomic/ molecular weight	Melting point (°C)	Boiling point (°C)	Density (g/cm^3)	Crystal system	CAS Number
Arsanilic acid	217.07	232	---	1.9571^{20}	monoclinic needles from water or ethanol	98-50-0
Arsenic	74.92	817 (28 atm) (triple-point)	613 (sublimes)	5.727^{14}	hexagonal, rhombic	7440-38-2
Arsenic pentoxide	229.84	315 (dec.)	---	4.32	amorphous	1303-28-2
Arsenic sulphide	246.04	300	707	3.43	yellow or red monoclinic needles (change from yellow to red at ~170°C)	1303-33-9
Arsenic trioxide[a]	197.84	312.3	465	3.738	amorphous or vitreous	1327-53-3
Arsine	77.95	-113.5	-55 (dec. 230)	2.695 (gas)[b] 1.689^{-49} (liq.)	colourless gas	7784-42-1
Calcium arsenate	398.08	---	---	3.62	amorphic powder	7778-44-1
Dimethylarsinic acid	138.00	200	---	---	prism	75-60-5
Lead arsenate	347.12	720 (dec.)	---	5.79	monoclinic leaves	7784-40-9
Methanearsonic acid, disodium salt	183.9	132-139	---	---	crystalline	144-21-8
Methanearsonic acid, monosodium salt	161.9	115-119	---	---	crystalline	2163-80-6
Potassium arsenate	180.04	288	---	2.867	tetrahedral	7784-41-0
Potassium arsenite	254.8	---	---	---	powder	13464-35-2
Sodium arsenate dodecahydrate	423.93	86.6	---	1.752-1.804	trigonal or hexagonal prism	7631-89-2
Sodium arsenite	129.91	---	---	1.87	powder	7784-46-5
Sodium cacodylate	159.98		200	---	crystalline	124-65-2

[a] Vapour pressure 0.653 (200°C)

[b] Specific gravity (air = 1)

Source: Adapted from International Agency for Research on Cancer (1980).

Arsenic pentoxide, As_2O_5, may be readily prepared by nitric acid oxidation of elemental arsenic or the trioxide. Compared to the trioxide, the pentoxide has considerable solubility in water (63.0 g/100 g water), presumably dissolving to form the relatively strong arsenic acid, H_3AsO_4. In acid media, arsenic acid has oxidizing potential (E°=0.56V).

The relative stability of solutions of arsenic or arsenous acids to oxidation-reduction is of considerable importance in terms of valency-dependent arsenic toxicity. In oxygenated media, one would expect the pentavalent form to dominate, while reducing media would favor the trivalent state.

Arsenous and arsenic acids both form mono-, di- and tri-metal salts, the alkali-metal salts such as potassium and sodium arsenite being freely soluble in water and the alkaline salts such as calcium or magnesium arsenite being slightly soluble.

While tri-organic esters of the tri- and pentavalent arsenic acids are known, they are labile to hydrolysis and one would expect the mono- and di-organic derivatives to be even more so. This behavior has implications in the postulated role of arsenate ions in interfering with phosphorylation reactions.

Arsine (arsenic trihydride, AsH_3) is a strong hemolytic toxicant and probably the most poisonous of the arsenicals. Although generally a minor factor in the environmental chemistry of arsenicals, it can form under certain restricted conditions, i.e., via reduction in the presence of an active hydrogen-generating source.

Monomethyl and dimethyl arsenic, in the form of methylarsonous and methyl arsonic, dimethylarsinous and dimethylarsinic (cacodylic) acids, occur both in the environment and are formed via in vivo transformation in many mammalian species, including man. Such organic arsenic compounds are also of commercial significance. For instance, both methylarsonic and dimethylarsinic acids, usually in the form of the mono- or dialkali salts, are employed as herbicides which, when released into the environment, may undergo reduction to the corresponding labile arsine compounds, CH_3AsH_2 (methylarsine) and $(CH_3)_2$ AsH (dimethylarsine) (Arsenic. NAS, 1977). Like trivalent inorganic arsenic, methylarsonous acid can interact with thiol groups (as can cacodylic acid) to form the CH_3-As(-S-)$_2$ and $(CH_3)_2$-As-S groups, respectively. The physical forms of arsenic in the environment depend on its mode of emission and subsequent interactions with other materials.

Arsenic, along with other trace metals, can be mobilized in association with airborne particles derived from high-temperature sources such as fossil-fueled power plants, metallurgical smelters and blast furnaces.

Arsenic compounds form insoluble complexes with soils and sediments. With soils, the interaction involves clay surfaces containing amorphous aluminum or iron oxides (Woolson, 1976).

3.2 ENVIRONMENTAL CYCLING OF ARSENIC

Inorganic arsenic is released into the environment from a number of anthropogenic sources which include primary copper, zinc and lead smelters, glass manufacturers (specifically those that add arsenic to the raw materials) and arsenic chemical manufacturers. Figure 3-1 presents a generalized scheme for the geochemical cycling of arsenic through various compartments of the environment. The atmosphere is a major conduit for arsenic emitted from anthropogenic sources to the balance of the cycle via wet and dry precipitation processes. The rate of movement of arsenic from the atmosphere is not known at present.

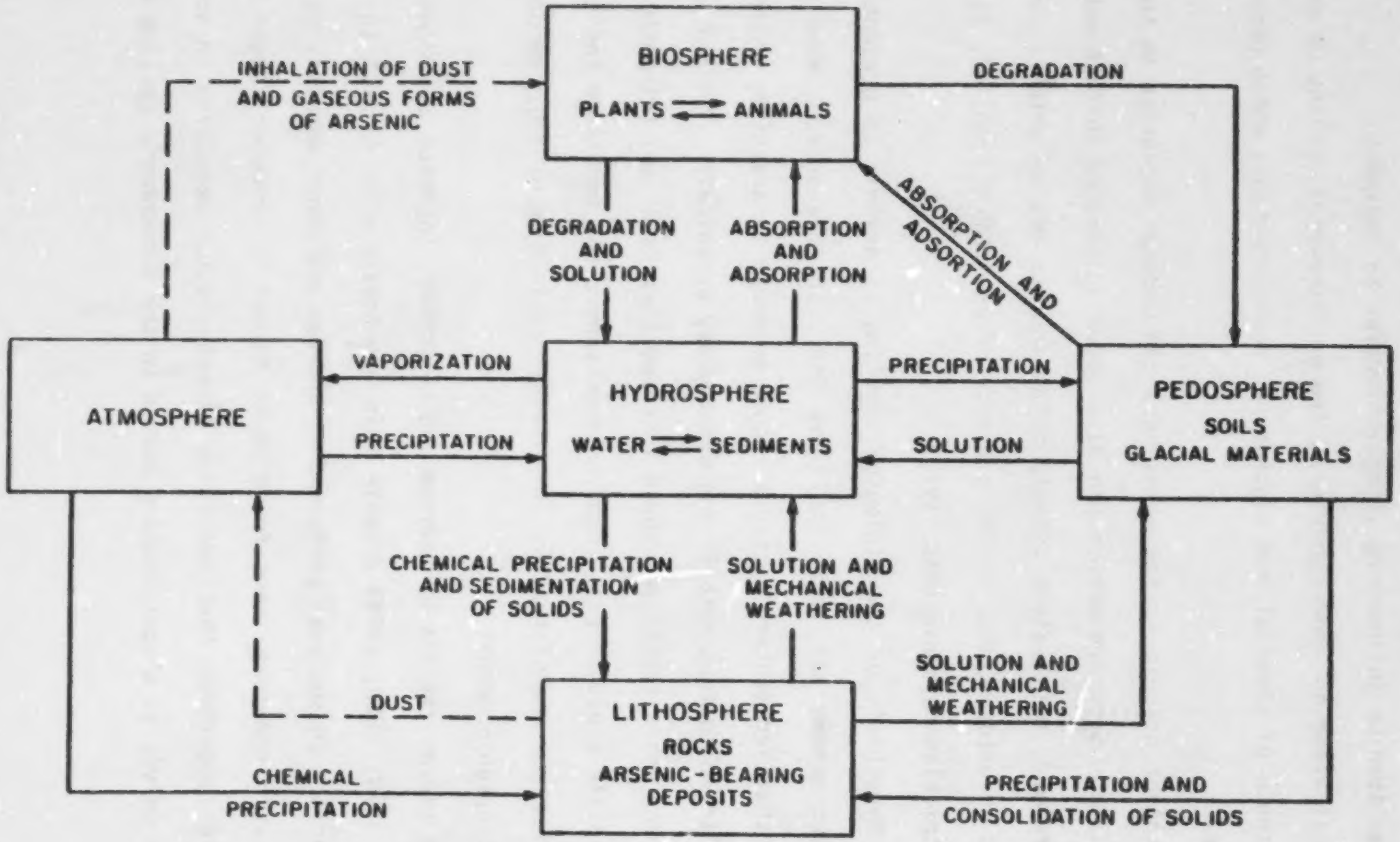

INHALATION OF DUST AND GASEOUS FORMS OF ARSENIC
BIOSPHERE
PLANTS ⇄ ANIMALS
DEGRADATION
DEGRADATION AND SOLUTION
ABSORPTION AND ADSORPTION
ABSORPTION AND ADSORTION
ATMOSPHERE
VAPORIZATION
PRECIPITATION
HYDROSPHERE
WATER ⇄ SEDIMENTS
PRECIPITATION
SOLUTION
PEDOSPHERE
SOILS
GLACIAL MATERIALS
CHEMICAL PRECIPITATION AND SEDIMENTATION OF SOLIDS
SOLUTION AND MECHANICAL WEATHERING
SOLUTION AND MECHANICAL WEATHERING
DUST
LITHOSPHERE
ROCKS
ARSENIC-BEARING DEPOSITS
CHEMICAL PRECIPITATION
PRECIPITATION AND CONSOLIDATION OF SOLIDS

Figure 3-1. The generalized geochemical cycle for arsenic.

Source: Union Carbide (1977).

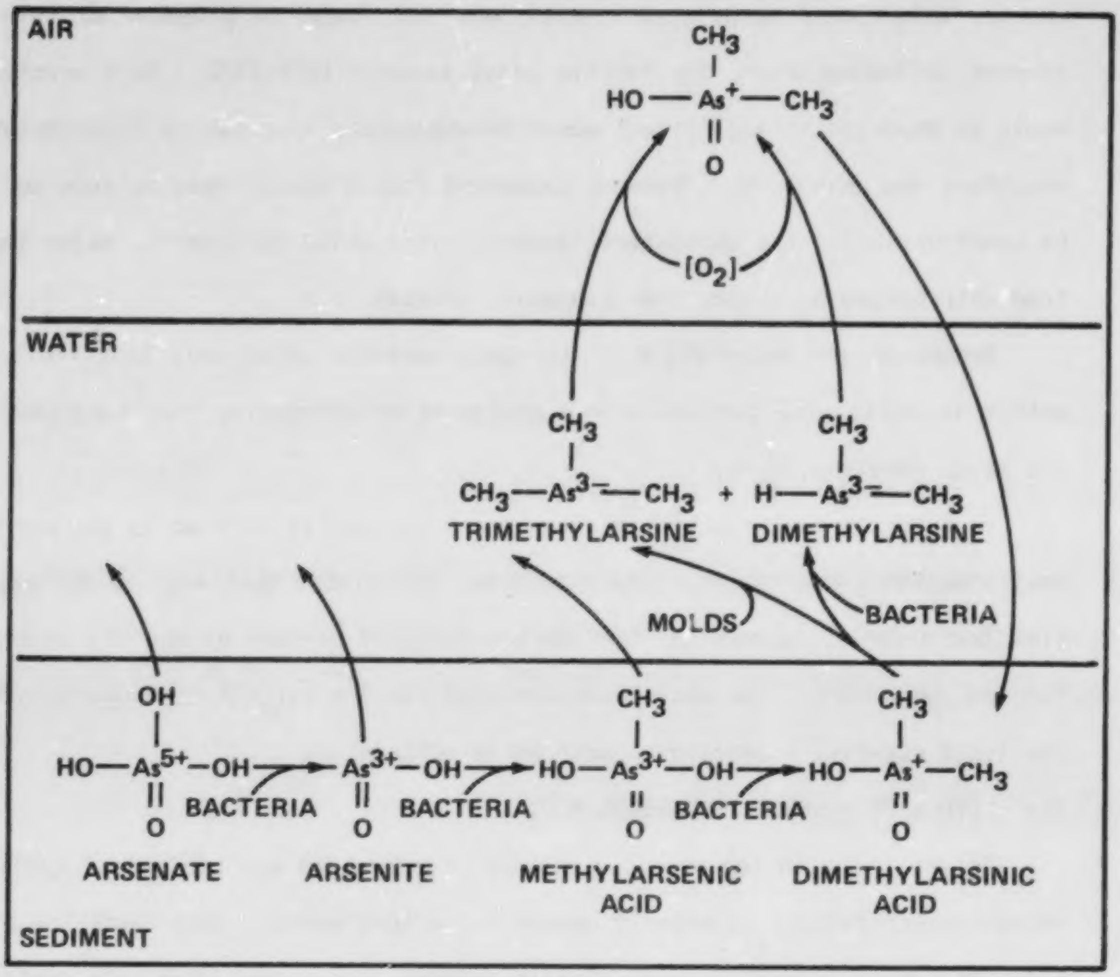
Dry and wet fall onto soils may be followed by movement through soils either into ground water or surface water. Passage of arsenic into surface waters may then be followed by further transfer to sediments.

Complicating an understanding of the environmental cycling of arsenic is the existence of chemical and biochemical transformations which occur within the cycle.

Trivalent arsenic in the atmosphere can undergo oxidation to the pentavalent state. Such conversion can also occur in aerated surface waters. On the other hand, pentavalent arsenic in an aqueous medium which is somewhat acidic is an oxidant, and, in the presence of oxidizable material, it will react to form trivalent arsenic (NAS, 1977).

One hypothesis of the biological cycling of arsenic is presented in a generalized scheme set forth in Figure 3-2. In this scheme, Wood (1974) proposes that sedimentary bacteria reduce arsenate to arsenite, which in turn may be altered to form methyl- and dimethyloxy arsenicals. Further action of bacteria and molds transforms these intermediates to di-and tri-methyl arsine. These volatile products pass into the hydrosphere and then into the atmosphere, where oxidative transformation to dimethyl-arsinic acid and, further, to inorganic arsenic occurs.

More recent reports by Andreae and co-workers (Andreae and Klumpp, 1979; Andreae, 1979; 1980; 1983) dispute this hypothesis with respect to marine environments. In analyses performed by Andreae and other workers, no volatile organoarsenic compounds were found to be formed in seawater, negating the hypothesis suggesting that reductive biomethylation resulting in volatile compounds serves as a contributing source to the atmospheric cycling of arsenic.



AIR
CH3
HO—As+—CH3
O
[O2]
WATER
CH3
CH3—As3−—CH3
TRIMETHYLARSINE
+
CH3
H—As3−—CH3
DIMETHYLARSINE
MOLDS
BACTERIA
OH
HO—As5+—OH
O
ARSENATE
BACTERIA
As3+—OH
O
ARSENITE
BACTERIA
CH3
HO—As3+—OH
O
METHYLARSENIC ACID
BACTERIA
CH3
HO—As+—CH3
O
DIMETHYLARSINIC ACID
SEDIMENT

Figure 3-2. The proposed biological cycle for arsenic.

Source: Wood (1974)

While the authors did note the occurrence of biomethylation, the observed products were non-volatile, water-soluble methylated oxoacids of low toxicity. Further, the site of biotransformation was reported to be planktonic algae residing in the photic zone, rather than sedimentary bacteria. Similarly, the authors noted that methylated species were not found in a number of rain samples collected along the Pacific coast between 1976-1978. Such species would be expected if significant ocean-to-atmosphere transfer by biomethylation reactions was occurring. Andreae suggested that a global mass balance could be constructed for the atmospheric arsenic cycle which derived its major input from anthropogenic, rather than biogenic, sources.

Reduction and methylation of inorganic arsenic occur only to a limited extent in soils, 1-2 percent over a period of months having been reported in one study (Woolson, 1976).

In terms of the relative amounts of arsenic partitioned among the various environmental compartments, Suta (1980) has calculated that land is the major sink for arsenic, accounting for approximately 90 percent of the dissipation for the year 1974. The atmosphere accounts for 7-8 percent dissipation with the least quantity appearing in waterborne effluents.

3.3 LEVELS OF ARSENIC IN VARIOUS MEDIA

As was noted in the previous section, geochemical and biological cycling mechanisms contribute to arsenic burden in various media. Such burdening of a given medium is augmented from specific sources, e.g., agro chemicals containing arsenic applied to agricultural lands.

Available information on the arsenic content of media with which man interacts is generally in the form of total amounts of the element, with limited data available for assessing specific chemical forms. Since the toxicity of arsenicals varies with chemical form, any supporting data for determining chemical states will be discussed in this report, however fragmentary.

3.3.1 Levels of Arsenic in Ambient Air

The most comprehensive data for ambient air levels of arsenic in the U.S. are those of the National Air Sampling Network conducted by the U.S. EPA. Measurements from this network are summarized in Table 3-2. All values were determined from nuclear activation analysis of individual 24-hour high volume particulate samples. As shown in Table 3-2, the number of observations available is considerably different from year to year. This is due to changes in the participation of state and local agencies operating the individual sites and, for the year 1981, only about half the samples collected have been analyzed and reported into the network data base at this time. Thus, direct comparison of the summary statistics from year to year may be of limited utility.

A closer examination of the NASN data by site indicates that in areas not influenced by copper smelters, maximum 24-hour concentrations do not exceed 0.1 μg/m^3. There are only two exceptions of approximately 600 site/years represented in the table where there were observations above 0.1 μg/m^3--Omaha, Nebraska, and Charleston, West Virginia. The site in Charleston ceased operation after 1978; therefore, it is difficult to interpret the single value above 0.1 μg/m^3. Repeated observations above 0.1 μg/m^3 have occurred at the site in Omaha, Nebraska.

Using the NASN data base, U.S. annual arithmetic mean arsenic concentrations are plotted in Figure 3-3. The plot suggests considerable variability over the five-year period with no clear direction of trend. Perhaps the most striking feature in the data is the relatively low annual arsenic level observed in 1980. A partial explanation for this observation may be the strike which idled most of the nation's copper smelters from June through September of 1980 (Eldred et al., 1983).

TABLE 3-2. CUMULATIVE FREQUENCY DISTRIBUTION OF NASN INDIVIDUAL 24-HOUR AMBIENT AIR ARSENIC LEVELS

Year	Minimum Detection Limit	Number of Observations	Percentile[a]				Arithmetic	
			30	50	70	99	Mean	(SD)
1977	.004	5385	0	0	.004	.048	.0049	.0165
1978	.006	1679	.006	.006	.008	.075	.0109	.0253
1979	.005	1263	.005	.005	.005	.077	.0091	.0192
1980	.007	2934	0	0	0	.037	.0026	.0113
1981	.007	688	0	0	0	.058	.0059	.0275

[a] Percentile values indicate the percentage of stations below the given air level.

Values in μg/m^3.

Source: National Arsenic Data Base, OAQPS/OANR, U.S. Environmental Protection Agency (Akland, 1983).

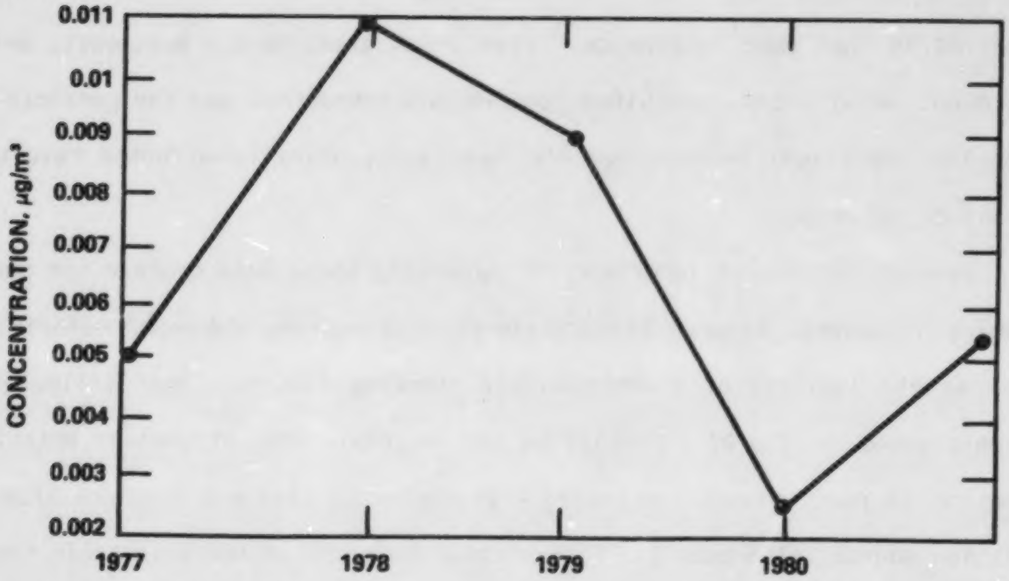


Figure 3-3. NASN annual average arsenic concentrations.

Source: National Atmospheric Data Base, OAQPS/OANR, U.S. Environmental Protection Agency (Akland, 1983).

The locations of these primary copper smelters in the U.S. are listed in Table 3-3. Examination of the NASN data by individual sites shows that the highest arsenic levels are consistently recorded in areas of Arizona, Montana, Tennessee, Texas, and Washington, which are impacted by the copper smelting industry. Thus, the closure of many of these plants for a substantial period in 1980 may have had the effect of depressing the nationwide average concentration of arsenic in ambient air for that year. Unfortunately, the possible effect of the 1980 copper smelter strike cannot explain the low arsenic levels observed in that year nationwide. Even urban areas in the Northeast, which would not be affected, exhibited lowered concentrations and the possibility that the analytical methodology may have systematically affected results cannot be ruled out.

Several factors of importance in assessing these data concern the partitioning of arsenic between particulate-bound fractions and vapor material as well as the lability of arsenic on air sampling filters. Non-particulate arsenic would chiefly be a problem in the immediate area of smelter emissions, adhesion to particulates increasing with residence time and distance from the emission source. Thompson (1976) has noted that all of the arsenic in the air in regions of smelters is adhered to particulates when sampled 2-3 kilometers from the operation. Lao et al. (1974) have cautioned about the hazard of arsenic loss, chiefly as the trioxide, from samplers at levels of 1.0 microgram or less. Walsh et al. (1977a), however, could not remobilize particulate arsenic once trapped in samplers. Whatever the hazard, the NASN values are internally consistent, remote areas having the lowest values and regions of smelter operations having the highest levels.

An important consideration in assessing the significance of air exposure of arsenic to health risk is the chemical form or forms of arsenic in the

TABLE 3-3. PRIMARY COPPER SMELTERS IN THE UNITED STATES

	Location	Company
1.	Anaconda, Montana	Anaconda Company
2.	Tacoma, Washington	Asarco, Inc.
3.	Garfield, Utah	Kennecott Copper Corp.
4.	El Paso, Texas	Asarco, Inc.
5.	McGill, Nevada	Kennecott Copper Corp.
6.	Hidalgo, New Mexico	Phelps Dodge Corp.
7.	Hurley, New Mexico	Kennecott Copper Corp.
8.	Hayden, Arizona	Kennecott Copper Corp.
9.	Hayden, Arizona	Asarco, Inc.
10.	Miami, Arizona	Inspiration Consolidated Copper Corp.
11.	Morenci, Arizona	Phelps Dodge Corp.
12.	Ajo, Arizona	Phelps Dodge Corp.
13.	Douglas, Arizona	Phelps Dodge Corp.
14.	San Manuel, Arizona	Magma Copper Company
15.	Copper Hill, Tennessee	Cities Services Company
16.	White Pine, Michigan	Copper Range Company

Source: National Arsenic Data Base, OAQPS/OANR, U.S. Environmental Protection Agency (Akland, 1983).

ambient air of the United States. Work by Johnson and Braman (1975), as well as that of Attrep and Anirudhan (1977), indicates that methylated arsenic, possibly in significant amounts, has been found in air samples, but its presence can likely be ascribed to either biotic activity or the use of methylated arsenics as herbicides. This is not in total agreement with Andreae (1983), who suggests that methylated atmospheric arsenic is due predominantly to anthropogenic sources, based upon his studies of marine environments. Methylated forms become a minor factor in suburban and urban areas.

It is not clear from the available data what forms of inorganic arsenic are in most air samples: trivalent, pentavalent or mixtures of these two oxidation forms. Crecelius (1974) found that rain water samples for an urban area of the western U.S. contained only about one-third (35 percent) trivalent arsenic. In a study conducted at various sites along the Pacific coast, Andreae (1980) observed wide variations in the ratio of arsenite to arsenate in rain. He attributed this variability to emissions of predominantly pentavalent arsenic from the sea surface and trivalent arsenic from industrial emissions (particularly those from a copper smelter in the Northwest), and by redox reactions during the residence of arsenic in atmospheric particulates and hydrometeors. Caution should be exercised, however, in interpreting any extrapolations from washout samples to original air composition, given the lability of inorganic arsenic to oxidize or reduce depending on aeration, acidity of the rain sample, and presence of oxidizable matter.

In summary, then, one can assume that methylated arsenic is of minor concern in suburban and urban/industrial air samples, and that the major inorganic portion is a variable mixture of the trivalent and pentavalent forms.

3.3.2 Levels of Arsenic in Drinking Water

The National Interim Primary Drinking Water Regulations, promulgated under the Safe Drinking Water Act, set the Maximum Contaminant Level (MCL) for arsenic in U.S. public water supplies at 50 μg/l (40 CFR Part 141). EPA is currently reviewing the arsenic MCL as part of its program to promulgate National Revised Primary Drinking Water Regulations (EPA, 1983).

In general, arsenic is not found in drinking water at levels exceeding the MCL. The Community Water Supply Survey conducted in 1969 found only 2 of the 969 supplies examined (0.2 percent) with levels exceeding 50 μg/l (McCabe et al., 1970). More recently, the Rural Water Survey conducted in 1978 reported that 0.8 percent of rural households exceeded the 50 μg/l level. Small rural communities showed a much higher population (6.6 percent) of households exceeding 50 μg/l than large rural communities (0.0 percent) and other rural areas (0.4 percent). The proportion of households exceeding 50 μg/l was greatest in the West (2.1 percent) and North Central (1.8 percent); none were found to exceed 50 μg/l in the Northeast or South (Francis et al., 1982). Compliance monitoring of public water supplies as required by the current drinking water regulations has revealed that approximately 100 of the nation's 60,000 public water supplies (0.2 percent) have levels exceeding the current MCL (Wentworth, 1983).

Notable exceptions to the generally favorable picture for U.S. population exposure to drinking water arsenic are isolated well water sources associated with geochemical enrichment by arsenic found mainly in the western U.S. and Alaska (Arsenic. NAS, 1977; Whanger et al., 1977; Harrington et al., 1978; Southwick et al., 1981). Whanger et al. (1977) have noted that well water arsenic levels in Lane County, Oregon, have ranged up to 2.2 ppm (one well) with levels generally increasing with well depth. Harrington et al. (1978) also noted that an area of Fairbanks, Alaska, had well water levels ranging as high as 10 ppm, representing geochemical input as well as contamination by residues from prior gold-mining activity. Southwick et al. (1981) reported levels ranging from 0.18-0.27 ppm in desert communities in Utah.

As with arsenic in air, it is important to take into account available information regarding the chemical forms of arsenic in the potable water supplies. It is reasonable to assume that the chief form of arsenic in most municipal water supplies, particularly surface reservoirs, would be pentavalent arsenic due to aeration and chlorination. Similarly, the major forms of arsenic in well waters relatively enriched in arsenic have been analyzed and also appear to be predominantly in the pentavalent inorganic state (Whanger et al., 1977; Harrington et al., 1978; Southwick et al., 1981). The chemical character of arsenic-rich well water in the U.S. versus other regions of the world will be taken up elsewhere.

3.3.3 Arsenic in Food

Perhaps the most useful body of data for assessment of the contribution of food arsenic to the total exposure picture in the U.S. is the recent survey carried out from August 1975 to July 1976 by the U.S. Food and Drug Administration (Johnson et al., 1981a,b). In Table 3-4 are summarized the average arsenic levels in various adult food classes for the period noted above; values are expressed analytically as arsenic trioxide. Corresponding average arsenic levels in food classes of 6-month-old infants and 2-year-old toddlers are summarized in Table 3-5.

In comparing the mean levels in comparable food categories, it appears that for children and adults, meat, fish, and poultry constitute the greatest dietary source of arsenic. Within this category, shellfish and other marine foods contain the highest levels of arsenic (Jelenik and Corneliussen, 1977). For infants, grain and cereal products constitute the only known source of arsenic intake from the categories measured.

TABLE 3-4. LEVELS OF ARSENIC (As_2O_3) BY FOOD CLASS IN ADULT FOOD COMPOSITES FROM 20 U.S. CITIES (AUGUST, 1975 - JULY, 1976)

Food Class Composite	No. of Positive Composites	As, ppm Mean (Range)
Dairy products	1	0.004(0.08)[a]
Meat, fish and poultry	17	0.19(0.03-0.46)
Grain and cereal products	8	0.02(0.03-0.10)
Potatoes	ND[b]	----
Leafy vegetables	ND	----
Legume vegetables	1	0.004(0.07)
Root vegetables	1	0.004(0.07)
Garden fruits	1	0.005(0.10)
Fruits	ND	----
Oils, fats and shortening	1	0.002(0.04)
Sugar and adjuncts	ND	----
Beverages (including drinking water)	1	0.008(0.15)

[a] Mean values are based on 20 composites for every food class.

[b] ND = not detected

Source: Adapted from Johnson et al. (1981a).

TABLE 3-5. LEVELS OF ARSENIC (As_2O_3) BY FOOD CLASS IN INFANT AND TODDLER FOOD COMPOSITES FROM 10 U.S. CITIES (AUGUST, 1975 - JULY, 1976)

Food Class Composite	No. of Positive Composites	As, ppm Mean (Range)	
		Infant	Toddler
Water	ND[a]	----	----
Whole milk, fresh	ND	----	----
Other dairy and substitutions	ND	----	----
Meat, fish and poultry	ND(I) 7(T)	----	0.092 (0.06-0.29)[b]
Grain and cereal products	3(I) 2(T)	0.018(0.04-0.09)	0.008(0.03-0.05)
Potatoes	ND	----	----
Vegetables	ND	----	----
Fruits and fruit juices	ND	----	----
Oils and fats	ND(I) 1(T)	----	0.003(0.03)
Sugar and adjuncts	ND(I) 1(T)	----	0.004(0.04)
Beverages	ND	----	----

[a] ND = not detected

[b] Mean values are based on 10 composites for every food class.

Source: Adapted from Johnson et al. (1981a).

Johnson et al. (1981a) have calculated that the total adult daily intake of arsenic (as As_2O_3) for August 1975 to July 1976 was approximately 65 µg. (Corresponding elemental content can be obtained by multiplying by 0.75. Thus, the intake of elemental arsenic would be 50 µg.) This represents an increase over the 1974 value of 21 µg reported by Jelenik and Corneliussen (1977). Differences in the two years can primarily be accounted for by increases in the arsenic content of meat, fish, and poultry; grains and cereal products; beverages; and dairy products. Johnson et al. (1981a) did not discuss whether the apparent increase in arsenic levels in certain categories represented a trend or merely reflected analytical variation from year to year. It should be noted, however, that arsenic mean levels in all food categories generally declined over the years 1967-1974. Therefore, data collected over the past few years (1977-present) but, as yet, unanalyzed, should be closely examined in order to determine whether the increases seen in 1975-1976 have actually constituted the beginning of a new trend.

In no other medium is the issue of chemical forms of arsenic more complex and simultaneously more important than in food, given the fact that the diet is a major, if not the main, source of arsenic for most of the U.S. population. As will be pointed out in a later section, arsenic in crustaceans and other marine life is present in the form of various chemically complex organoarsenicals, which appear to be not only resistant to metabolism but are rapidly excreted intact. Comparatively speaking, then, these forms are regarded as being toxicologically inert.

3.3.4 Arsenic in Soils

Soil arsenic levels are mainly of concern in this section to the extent that arsenic mobility and transformation in this medium allow for passage of the element to ground water, air and the food chain (via plant uptake). This area has been reviewed in some detail (Arsenic. NAS, 1977; Woolson, 1977; Walsh et al., 1977b).

Background levels of arsenic in soils range from less than 1 ppm to above 40 ppm, and the relative enrichment of this background level with agricultural practices is secondary to fallout from air in the regions of industrial activity and can be of the order of 100 times. In Table 3-6 are listed some comparative values for uncontaminated soils versus soils contaminated by the repeated use of defoliants and insecticides containing arsenic (Walsh and Keeney, 1975).

Arsenic in soils is usually bound to clay surfaces containing amorphous aluminum or iron oxides, the degree of immobilization (adsorption) being a function of soil type, soil pH, phosphate levels, levels of iron or aluminum as well as residence in soil. Soil-adsorbed arsenic is rather inert to transformation or movement and it is the soluble fraction which is of interest, usually in the pentavalent form. Pentavalent arsenic arises from both soil aeration and degradation of methylated arsenic herbicides. Little reductive methylation of arsenic occurs in typical soils compared to sediments, Woolson (1976) observing that only 1-2 percent conversion of arsenate occurred over a period of months.

Transfer of soil arsenic to plants entails the soluble, labile fraction of arsenic and the site of uptake is the root system where highest levels are found. Edible portions of most food plant classes are low. This is also the case for terrestrial flora, but marine plant life such as algae accumulates considerable levels of arsenic (Irgolic et al., 1977).

3.3.5 Other Sources of Arsenic

Small and McCants (1962) found an average of 1.5 ppm arsenic in tobacco residues taken from U.S. tobacco grown in soils having average arsenic levels

TABLE 3-6. A COMPARISON OF ARSENIC LEVELS IN ARSENIC-TREATED AND UNCONTAMINATED SOILS IN NORTH AMERICA

Sampling site	Total As content, ppm uncontaminated soil	Total As content, ppm treated soil[a]	Crop
Colorado	1.3 - 2.3	13 - 69	Orchard
Florida	8	18 - 28	Potato
Idaho	0 - 10	138-204	Orchard
Indiana	2 - 4	56 - 250	Orchard
Maine	9	10 - 40	Blueberry
Maryland	19 - 41	21 - 238	Orchard
New Jersey	10.0	92 - 270	Orchard
New York	3 - 12	90 - 625	Orchard
North Carolina	4	1 - 5	Tobacco
Nova Scotia	0 - 7.9	10 - 124	Orchard
Ontario	1.1 - 8.6	10 - 121	Orchard
Oregon	2.9 - 14.0	17 - 439	Orchard
	3 - 32	4 - 103	Orchard
Washington	6 - 13	106 - 830	Orchard
	8 - 80	106 - 2553	Orchard
	4 - 13	48	Orchard
Wisconsin	2.2	6 - 26	Potato

[a] These are results from soils that had been repeatedly treated with an As pesticide or defoliant. Soils treated experimentally are not included.

Source: Walsh and Keeney (1975).

of 3 ppm. Of the total arsenic content of cigarette tobacco, 10-15 percent is in mainstream smoke (Thomas and Collier, 1945) in an unidentified form. In the past, levels of up to 40 ppm arsenic were detected in U.S. cigarettes (Holland and Acevedo, 1966) owing to arsenical use as pesticides. Lower current levels probably reflect curtailed use of arsenic as a pesticide in tobacco growing.

While current levels of arsenic may be lower in tobacco than had been the case, past exposure of cigarette smokers remains a health factor in considering respiratory cancer risk, given the long clinical induction period (decades) for this health effect (see Chapter 7).

4. ARSENIC METABOLISM

4.1 ROUTES OF ARSENIC ABSORPTION

The significance of various routes of arsenical intake for man and various other animal species is dependent upon the physical and chemical form of the arsenical, the mode of exposure, and the animal species under study. The major routes that are of significance to general public health are inhalation and ingestion, either via direct intake of food and water or secondary intake via the inhalation of arsenic in a form and size where it is eventually swallowed. Inhalation is probably of more significance in occupational settings, while oral intake is the major exposure route for the population at large.

Percutaneous absorption of arsenic, while poorly studied, appears to be a relatively minor route of exposure except under certain occupational exposure conditions.

4.1.1 Respiratory Absorption

Some quantitative and qualitative information about the respiratory deposition and absorption of arsenic by human subjects has been reported. Holland and co-workers (1959) used a group of hospital patients (lung cancer) to assess the deposition and absorption of inhaled arsenic using arsenite-containing cigarettes labeled with arsenic-74 as well as arsenite-containing aerosols. Deposition amounted to approximately 40 percent and more than three-fourths (75 to 85 percent) of the deposited arsenic was absorbed from the lungs within 4 days. While it may be argued that the health status of these subjects may have influenced the extent of absorption, it is nevertheless reasonable to infer that relatively rapid and extensive absorption of arsenic from the human lung likely occurs.

A study of a group of workers (Pinto et al., 1976) exposed to airborne arsenic in a copper smelter in 5-day test periods demonstrated average urinary arsenic excretion values ranging from 38 to 539 μg/liter to be associated with averages of air levels of arsenic ranging from 3 to 295 $\mu g/m^3$ (overall 53 μg As/m^3). Urinary arsenic was correlated with workplace air exposure (0.53, $p < 0.01$). Either direct pulmonary absorption or swallowing of larger particulate matter was evident as seen from elevations of urinary arsenic within 24 hours.

In a later study of copper smelter workers in which urinary excretion of arsenic was determined as total levels and chemically variant forms, Smith et al. (1977) studied control subjects and individuals from low, medium, and high arsenic exposure groups. In that study, the variations in concentrations of all arsenic forms isolated in urine--trivalent, pentavalent, methyl, and dimethylarsenic--were directly correlated with levels of airborne exposure (the relevant issue of arsenic biotransformation in vivo being discussed elsewhere in this chapter). An important feature of the Smith et al. (1977) report was the observation of a difference in the tightness of the correlation between excretion and exposure as a function of particle size. Both smaller, clearly respirable (<5 μm) and larger, less respirable (>5 μm) arsenic particles correlated well with excretion levels, but the less respirable arsenic-excretion relationship was seen to be much stronger than that for the finer mode particles. According to the authors, this is due to the swallowing of the large particle fraction and significant absorption from the GI tract.

Some animal data have also been reported on arsenic absorption via the respiratory tract. Bencko and Symon (1970) observed that hairless mice breathing a solid aerosol of fly ash containing 180 μg As/m^3 for several weeks had increases in tissue arsenic values. Since the particle size was determined to be only less than 10 μm, part of this intake may have occurred via the GI tract. Increases in tissue arsenic in two exposure groups also occurred when rats were exposed to arsenic trioxide (condensation aerosols: 1.0, 3.7, and 46 $\mu g/m^3$) for 90 days (Rozenshtein, 1970).

Relatively rapid absorption of pentavalent arsenic (arsenate solution) was noted by Dutkiewicz (1977) when rats were exposed intratracheally (arsenate solution labeled with arsenic-74; 0.1 and 4.0 mg/kg). Arsenic tissue distribution dynamics were similar for intratracheal and companion intravenous exposure studies, indicating that the rate of intratracheal arsenic uptake more closely resembles that from parenteral administration than do oral or percutaneous exposures.

The pulmonary retention of arsenic compounds with different solubilities has recently been studied by two research groups.

Inamasu et al. (1982) gave 61 male Wistar rats single intratracheal instillations of arsenic trioxide or calcium arsenate suspended in phosphate buffer (pH 6.9). Controls (19) were given instillations of the phosphate buffer solution. The total dose of arsenic administered to each animal was about 2 mg in 0.2 ml suspensions.

Four to five of the arsenic-exposed animals were killed at intervals from 15 minutes to 168 hours after instillation. The average amounts of arsenic recovered in lungs of rats at 15 minutes after the instillation of calcium arsenate and arsenic trioxide were 1146 μg and 620 μg, respectively. After 24 hours, almost all of the deposited arsenic trioxide had disappeared from the lungs, whereas only about 50 percent of the calcium arsenate had been cleared as shown in Figure 4-1.

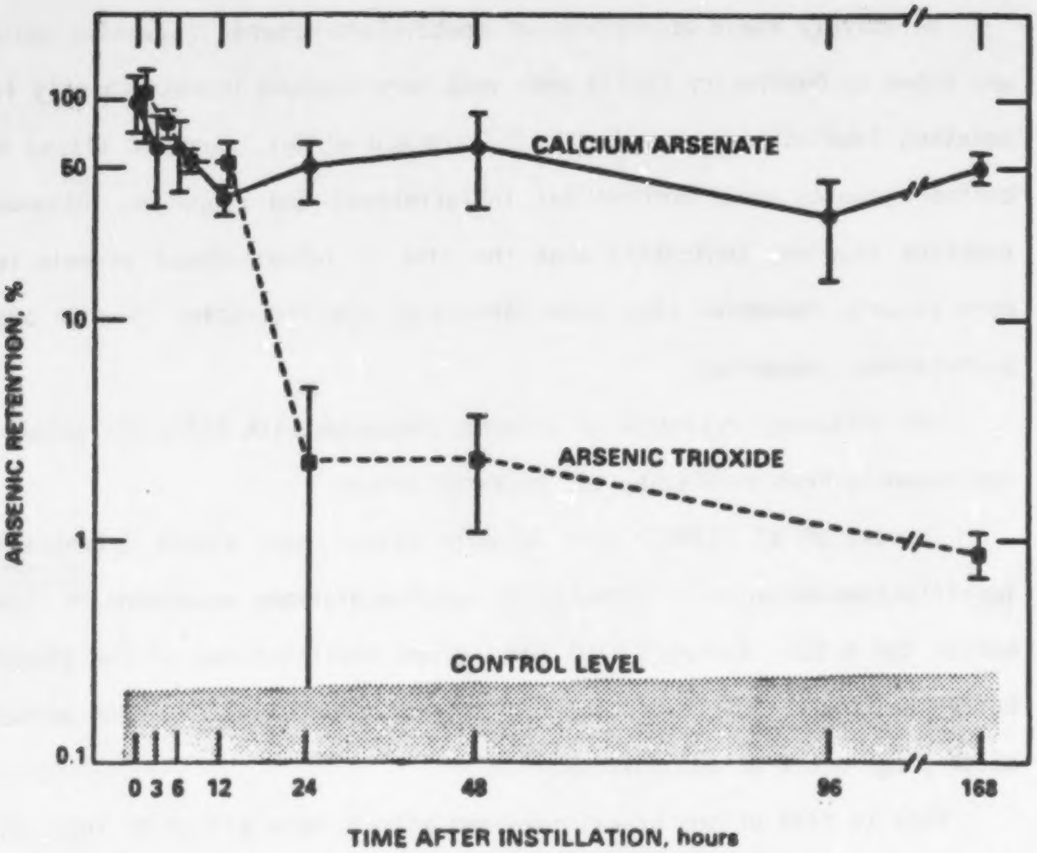


Figure 4-1. Arsenic retention in rat lungs following intratracheal instillation of a single dose. Percentage values are based on the average amount of arsenic present in rat lungs at 15 minutes after instillation of arsenic trioxide (- ● -) or calcium arsenate (—●—). Vertical bars: means ± S.D.

Source: Inamasu et al. (1982).

Of the small remaining amount of arsenic trioxide, most gradually disappeared over the next several days, declining to a level of 5.6 μg at 168 hours after instillation, a level slightly higher than that seen in the controls (1.3 μg). In contrast, calcium arsenate remained at about the same level 168 hours after instillation as was observed after 24 hours (768 μg), clearly demonstrating a much higher level of retention compared to arsenic trioxide.

In the study by Pershagen et al. (1982), male Syrian golden hamsters were given four weekly intratracheal instillations of suspensions of arsenic trioxide, arsenic trisulfide and calcium arsenate in doses of 0.3, 0.5 and 0.5 mg arsenic, respectively. The suspensions were made in 0.9 percent NaCl, and sulfuric acid was added accordingly to equalize the acidity of the three different suspensions. Twenty animals were assigned to each treatment group. Five animals in each group were killed immediately after the first instillation. Two to five animals were then killed one week after two or four instillations and two weeks after four instillations.

The mortality was highest among the animals exposed to arsenic trioxide. In this group, 9 of the remaining 11 animals not sacrificed for tissue analysis had died by the week following the third instillation. In contrast, at the second instillation, two animals died in the group receiving arsenic trisulfide but no further deaths occurred in this group. A total of three animals receiving calcium arsenate died during the first week following the fourth instillation. Aside from those animals specifically sacrificed for tissue analysis, no other deaths occurred in this group either.

In the lungs, the amounts of arsenic were 386, 755 and 866 mg/kg wet weight immediately following instillation of arsenic trioxide, arsenic sulfide and calcium arsenate, respectively. One week after the second instillation the amounts were 0.81, 9.2 and 579 mg/kg, respectively. In Figure 4-2 it can be seen that after the fourth instillation the differences between arsenate-exposed animals and the other groups were even more pronounced. Examination of the lungs revealed severe lung damage only among animals exposed to calcium arsenate. In all groups, areas with epithelial hyperplasia and metaplasia were seen.

Although different species and different time intervals were used in the above studies, the results were very consistent, i.e. arsenic trioxide was rapidly cleared from the lungs, whereas calcium arsenate was very slowly eliminated. The differences appear to be related to the relative solubility of the compounds, calcium arsenate having the lowest solubility. The doses in these two experiments were of the same magnitude, but the lung damage reported by Pershagen et al. among the arsenate-exposed animals might have influenced the clearance mechanisms. In addition, the possibility of higher acidity in the suspensions used by Pershagen et al. may also have had an influence.

The large differences in retention of arsenic compounds demonstrated in these two studies are of great interest in relation to the association between airborne arsenic and lung cancer. These differences in retention might explain the relatively high concentration of arsenic found in deceased smelter workers whose last exposure to arsenic occurred many years prior to death (Brune et al., 1980) (see Section 4.4). These workers were thought to be exposed to arsenic trioxide, but it may be that the retained fraction noted in the lungs was another arsenic compound of less solubility.

4.1.2 Gastrointestinal Absorption

In both man and experimental animals, factors which govern the absorption of arsenic from the gastrointestinal tract include the chemical form of the element, its physical characteristics, and dosing level. It can be stated

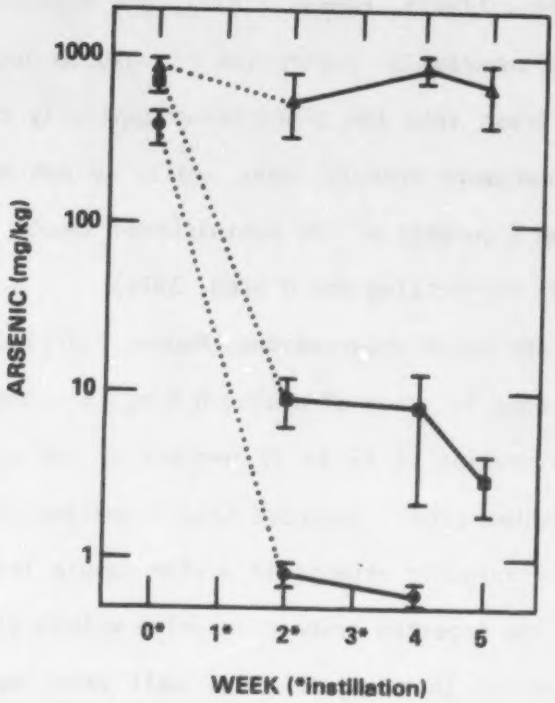


Figure 4-2. Lung concentrations of arsenic in hamsters given weekly intratracheal instillations of arsenic trioxide (●), arsenic trisulfide (■), or calcium arsenate (▲) (bars indicate ±1 S.D.). Animals at Week 0 were killed immediately following the first instillation; other animals were killed either 1 or 2 weeks after an instillation.

Source: Pershagen et al. (1982).

that soluble arsenicals will be generally more extensively absorbed than the insoluble forms. On the other hand, one should be cautious in extending correlations of simple water solubility to the chemical milieu existing in the GI tracts of various species.

Taken collectively, the reports of Coulson et al. (1935), Ray-Bettley and O'Shea (1975), Crecelius (1977), Mappes (1977), and Buchet et al. (1981a,b) demonstrate that very substantial absorption of soluble inorganic trivalent arsenic from the GI tract into the bloodstream typically occurs. Greater than 95 percent of inorganic arsenic taken orally by man appears to be absorbed, with less than 5 percent of the administered amount appearing in feces (Coulson et al., 1935; Ray-Bettley and O'Shea, 1975).

Consistent with the above observations, Mappes (1977) observed that daily oral intake of an aqueous solution of around 0.8 mg trivalent arsenic resulted in a daily urinary excretion of 69 to 72 percent of the daily dose in one subject. Also, Crecelius (1977) reported that ingestion of 50 μg trivalent and 13 μg pentavalent inorganic arsenic in a wine sample led to the appearance of 80 percent of all the ingested arsenic in urine within 61 hours. Crecelius (1977), however, reported that ingestion of well water mainly containing identified pentavalent inorganic arsenic led to urinary clearance of half the intake within approximately 3 days.

More recently, Buchet et al. (1981b) found that daily arsenic excretion via urine in human volunteers exposed to the dose range 125-1000 μg/day amounted to 60 percent of the dose. Steady state was achieved within 5 days after arsenic dosing began.

In contrast to the relatively high absorptive rate for soluble inorganic arsenic, Mappes (1977) reported that insoluble arsenic triselenide (As_2Se_3), when taken orally, passes through the GI tract with negligible absorption.

Arsenic intake via the diet of non-occupationally exposed populations requires that one consider the issue of bioavailability. Arsenic in foodstuffs is probably incorporated into the matrix of these commodities in a variety of ways.

The "shrimp" arsenic present in crustaceans and other fish appears to represent a complex organic form of the element which has prompted considerable recent study (LeBlanc and Jackson, 1973; Westöö and Rydälv, 1972; Munro, 1976; Edmonds et al., 1977; Penrose et al., 1977; Crecelius, 1977; Edmonds and Francesconi, 1977). In brief, the results of such studies indicate that the arsenic present in shellfish and other marine foods appears to be extensively absorbed and rapidly excreted intact as a complex organoarsenical by man and animals, and, as such, does not appear to pose a particular health threat to man.

Studies of the oral intake and absorption of arsenicals in experimental animals have also been conducted and generally confirm the findings derived from the above human studies. More specifically, soluble inorganic arsenic, delivered as either trivalent or pentavalent solutions, is almost completely absorbed from the GI tract of rats (Coulson et al., 1935), with 88 percent absorption being observed for arsenic trioxide solution (Urakabo et al., 1975; Dutkiewicz, 1977) and 70 to 90 percent for arsenate solution. Similar observations have been made for pigs (Munro et al., 1974), with 90 percent of arsenic trioxide solution being absorbed, and monkeys (Charbonneau et al., 1978a), with 98 percent of arsenic trioxide being absorbed. Also, Charbonneau et al. (1978b) fed arsenic-containing fish (Atlantic grey sole) to adult female monkeys as a homogenate (1 mg fish arsenic/kg body weight) and noted that about 90 percent of the intake was absorbed, of which about 75 percent appeared in urine after 14 days. In a related study, swine and adolescent monkeys were seen to absorb approximately 70 and 50 percent, respectively. On the other hand, arsenic trioxide in suspension given orally to rabbits and rats was reported to result in only about 40 and 30 percent absorption, respectively (Ariyoshi and Ikeda, 1974).

4.1.3 Transplacental Passage

Potential fetal exposure to toxic elements via the mother is a matter of major importance given the potential sensitivity of *in utero* development to deleterious impacts of exogenous toxic agents.

In an arsenic-detection study of maternal-newborn blood groups, Kagey et al. (1977) reported that cord blood levels approximated those of mothers in 101 subject sets. Tissue analysis (Kadowaki, 1960) of fetus arsenic in a presumably healthy Japanese population indicated measurable element levels by at least month four of gestation which increased to month seven. Of importance in this study was the observation that brain levels, as well as those of bone, liver, and skin, were the highest of all tissue tested.

Complicating the issue is the chemical nature of the tissue arsenic assayed in either of the two studies noted above, inasmuch as precise chemical speciation was not attempted. Also, the Japanese study presumably did not select material in a manner such that dietary histories could be discerned. Thus, questions can be raised regarding implications of these data for fetotoxicity.

Transplacental transfer of arsenic has also been demonstrated in experimental animals. Transplacental transfer of arsenate administered parenterally in hamsters has been demonstrated by Ferm's laboratory (Ferm, 1977; Hanlon and Ferm, 1977). The movement was rapid, with embryonic tissues showing levels close to those in maternal blood 24 hours after dosing. Trivalent arsenic exposure results in transplacental passage in pregnant rats. Arsenic was detected in newborn rats when the dams received arsenic trioxide in their diets.

4.2 BIOTRANSFORMATION PROCESSES IN VIVO

An understanding of the metabolism of inorganic arsenic by man and a number of other species is substantially complicated by a series of newly-revealed biotransformations, including methylation of inorganic arsenic. It is thus appropriate to discuss _in vivo_ transformation processes at this point, since many of the data dealing with blood transport, tissue distribution and subsequent excretion are much better understood in light of these findings.

A major factor in the determination of transformation processes for arsenic in man and other animals was the evolution of appropriate analytical methods within the decade which permit chemical speciation of chemically variant forms of arsenic with reference to both oxidation-state lability and inorganic versus organo-substituted arsenic.

These procedures involve different analytical approaches and include selective reduction of various arsenic forms to the hydride, and subsequent analysis by colorimetry (Lakso et al., 1979), emission spectrometry (Braman and Foreback, 1973; Crecelius, 1977), atomic absorption spectrometry (Edmonds and Francesconi, 1977; Buchet et al., 1980), and gas-liquid chromatography (Andreae, 1977; Talmi and Bostick, 1975). Alternatively, inorganic arsenic (III) or (V) and methylated forms can be directly measured by ion-exchange chromatographic techniques (Tam et al., 1978; Henry and Thorpe, 1980).

There are a number of points germane to consideration of _in vivo_ transformations of arsenic: 1) biomethylation of inorganic arsenic; 2) oxidation-reduction of inorganic arsenic _in vivo_; 3) the relative chemical stability of inorganic arsenic to oxidation-reduction as it affects experimental dosing and chemical speciation in biological media.

4.2.1 _Biomethylation of Inorganic Arsenic in Humans and Experimental Animals_

An extensive literature has recently appeared documenting the _in vivo_ methylation of inorganic arsenic to monomethyl and dimethyl arsenic (the latter being the major methylated metabolite) in every mammalian system studied to date, including man.

While the quantitative features of this phenomenon may vary among species, one can generally state that: 1) dimethyl arsenic is the major transformation product in both humans and animals, with higher amounts of monomethyl arsenic, when formed, found in humans; 2) methylation is assumed to be a route of detoxification of the more toxic inorganic forms; 3) dimethyl arsenic appears to be a terminal metabolite, formed relatively rapidly and rapidly excreted; 4) while the *in vivo* methylating capacity of a given system may persist over a range of inorganic arsenic exposure, at some point the body burden of the unmethylated fraction is enough to induce toxic effects, evidenced by the extensive literature dealing with inorganic arsenic toxicity; 5) retrospective assessment of earlier data dealing with arsenic metabolism, including distribution and excretion, must be viewed in light of current knowledge about biomethylation in different species; and 6) biomethylation appears to be a reductive process involving step-wise arsenic reduction and methyl transfer steps.

4.2.1.1 Human Studies--Using a method that permits the determination of tri- and pentavalent inorganic arsenic as well as mono- and dimethylarsenic acids via selective reduction, volatilization, and helium-arc emission detection, Braman and Foreback (1973) analyzed the urinary excretion of arsenic in four human subjects. About 66 percent of the total urine arsenic concentration (22.5 ppb) was present as dimethylarsinic acid and 17 percent as pentavalent inorganic arsenic. Trivalent inorganic arsenic and methylarsonic acid were present in equal amount, approximately eight percent each.

Crecelius (1977) reported the urinary excretion of form-variable arsenic when a human subject ingested arsenic-rich wine (50 μg trivalent and 13 μg pentavalent). About 80 percent of the arsenic ingested with the wine was excreted within 61 hours. Of the total excreted, 63 percent was in the form of dimethylarsenic acid, 18 percent was monomethyl arsenic acid, and approximately 9 percent each was in the two inorganic forms.

Consumption of well water containing 200 μg of arsenate by a subject in the same study showed urinary trivalent arsenic at near background levels with an elevation in pentavalent form as well as significant excretion of dimethylarsenic. Of the total amount ingested, about 50 percent was recovered in urine by 3 days. Arsenic as contained in canned crab tissue was also studied in this experiment. It has been established that arsenic is present in marine foods in an organic form which is excreted intact.

The study of Smith et al. (1977), using basically the same speciation/analysis techniques noted in the previous study and involving urinary profiles for a group of copper smelter workers, also confirmed transformation processes _in vivo_. In controls, as well as in three study groups that varied as to intensity of airborne trivalent arsenic oxide exposure, dimethyl arsenic was the dominant species in urine, followed by methyl arsenic, trivalent arsenic and pentavalent arsenic, in descending order.

In another smelter worker study, Buchet et al. (1980) assessed the distribution of arsenic and metabolites in urine samples in different groups according to the degree of inorganic arsenic exposure. Total urinary arsenic in exposure groups ranged from 74 to 934 μg/liter, of which 75 percent represented methylated forms (mono- and dimethyl), with dimethyl arsenic predominating. Control subjects showed a somewhat higher level of methylated arsenic in urine, approximately 90 percent, suggesting some dependency of the extent of methylation on the level of exposure.

A number of recent studies have described in more detail the biomethylation of inorganic arsenic using human volunteers.

Buchet et al. (1981a) followed the urinary excretion of inorganic and methylated arsenic in 3-5 volunteers ingesting single amounts of arsenic at a dose of 500 μg, in the form of either arsenite, monomethyl or dimethyl arsenic. After 96 hours, 46 percent of the dose in subjects given arsenite (trivalent arsenic) was excreted, 25 percent in the inorganic form and 75 percent as methylated metabolites. Of the latter, one-third was monomethyl arsenic and two-thirds was the dimethylated form. Dimethyl arsenic was excreted intact, 75 percent of the ingested amount appearing in urine by 96 hours. With monomethyl arsenic, excretion reached 80 percent by 96 hours, with around 13 percent of the urine level representing conversion to the dimethyl form.

In a follow-up report, these workers (Buchet et al., 1981b) monitored the urinary excretion of various arsenical forms in 4 human subjects undergoing repeated ingestion (5 days) of trivalent inorganic arsenic as a meta arsenite salt at 4 levels--125, 250, 500 or 1000 μg As. By 14 days, the percent of methylated forms of total forms ranged from 74-93 percent over the intake range. From this study, it would appear that the methylating capacity of the human subject was unaffected up to the 500-μg As level, at which point some decrease in methylated arsenic as a percentage of total was observed.

Yamauchi and Yamamura (1979) reported their results with 3 volunteer subjects who ingested an arsenic-rich extract (prepared from a particular kind of seaweed) containing 2.88 ppm total arsenic. Of the total amount, 86 percent was pentavalent inorganic arsenic, 7 percent was trivalent inorganic arsenic and 7 percent was dimethyl arsenic. These volunteers ingested the preparation as a single dose at a level of 10 μg/kg body weight, for total amounts of 650, 680, and 760 μg in the three subjects. By 48 hours, urine levels of total arsenic amounted to 36 percent of the dose. Of the excreted amount, dimethyl and monomethyl arsenic accounted for 47.4 and 25.3 percent, respectively.

4.2.1.2 Animal Studies--To date, biomethylation processes involving inorganic arsenic and experimental animals have been documented in dogs (Lakso and Peoples, 1975; Charbonneau et al., 1978a; Tam et al., 1979a), mice (Vahter, 1981), rabbits (Marafante et al., 1980; Bertolero et al., 1981), the bovine (Lakso and Peoples, 1975), and the rat (Odanaka et al., 1978). While the level of methylated forms of arsenic in most experimental species resembles that in man, approximately 80 percent, there is a greater amount of the dimethyl and a lesser amount of monomethyl arsenic.

Vahter (1981) has shown: (1) that the degree of biomethylation is dose-dependent, at least in mice, falling off in relative percentage with increasing level of dosing; (2) that this dependency probably accounts for the observation of dose-dependent retention in mice; and (3) methylation occurs to a greater extent with trivalent arsenic than with the pentavalent form, although retention, relative to total dose, is greater with the trivalent form.

Recently, Vahter et al. (1982) have reported that the Marmoset monkey, alone among all mammalian experimental species studied thus far, does not methylate inorganic arsenic.

4.2.2 In Vivo Oxidation/Reduction of Inorganic Arsenic In Mammalian Systems

From all available evidence, the biochemical mechanism of *in vivo* methylation of inorganic arsenic appears to be a reductive process, and sequential arsenic reduction and transmethylation steps in the pathway have been documented in microbial systems (Challenger, 1978; McBride et al., 1978). In mammalian systems, recent studies support a similar reduction/methylation sequence as observed in microflora.

Oxidation of trivalent inorganic arsenic to the pentavalent state has been claimed in dogs (Ginsberg, 1965), rats (Winkler, 1962), mice (Bencko et al., 1976), and humans (Mealey et al., 1959). An important factor in considering these studies critically is the fact that none of the reports considered the presence of methylated arsenic, which may well have affected the analytical techniques employed and the analytical data obtained. Secondly, except for the Bencko et al. (1976) study, it is not clear that careful attention was paid to the oxidation state composition of dosing media or the effect of sample handling on oxidation state stability. In the Mealey et al. (1959) report, the analytical method involved acidification of urine with hydrochloric acid and extraction of the presumed "trivalent" arsenic into benzene, leaving the presumed "pentavalent" form in the aqueous phase. Since Mushak et al. (1977) have shown that methylarsonic and dimethylarsinic (cacodylic) acid behave like the pentavalent inorganic form in this extraction procedure, it is probable that the "arsenate" fraction was mainly methylated arsenic.

In the study of Crecelius (1977), where chemical speciation techniques were employed, it does not appear that ingestion of a sample containing mainly arsenite (trivalent form) is associated with excretion of pentavalent inorganic arsenic as the chief inorganic form. After 61 hours, 80 percent of the ingested amount appeared in urine, with the two inorganic forms each constituting minor fractions, mono- and dimethyl arsenic being the main forms.

Evidence for the in vivo reduction of pentavalent to trivalent inorganic arsenic has been claimed in several early reports. Lanz et al. (1950) reported some reductive conversion (10-15 percent) of arsenate to arsenite in rats, using as a method the precipitation from urine of arsenate as a mixed salt and analysis of the supernatant for arsenic (III). The accuracy of this method for partitioning trace arsenic levels is questionable. Furthermore, at the time of this study, the presence of methylated arsenic was not known. Therefore, it is not possible to accept the authors' conclusions since unprecipitated arsenic could also have consisted of methylated forms.

Ginsberg (1965) reported that 14 percent of urinary and 6 percent of plasma arsenic in dogs receiving intravenous infusion of arsenate was in the form of trivalent arsenic. In this study, trivalent was presumably separated from pentavalent arsenic by chelation-extraction using ethyl xanthate. Based on current knowledge that methylated arsenic was present, it is difficult to accept the data without knowledge of the behavior of mono- and dimethyl arsenic in this analytical method.

In the recent report of Tam et al. (1979b) using dogs dosed with arsenate, a small amount of trivalent inorganic arsenic was detected in urine, using ion-exchange and thin-layer chromatographic techniques. These authors did not independently determine the extent of artifactive interconversion between the two forms using these techniques.

In a recent study by Vahter (1981), in which mice and rats were administered ^{74}As-labeled trivalent or pentavalent arsenic, the author reported that increases in retention with dose were less pronounced for the pentavalent form than for the trivalent form due to the fact that elimination of pentavalent arsenic seemed to be less dependent on methylation. Nevertheless, increases in retention with pentavalent arsenic were observed. Vahter discussed the possibility that the observed dose-related increase in retention after exposure to pentavalent arsenic may have been partly due to *in vivo* reduction of the pentavalent to trivalent form. Noting the work of McBride et al. (1978), whose studies indicated that certain microorganisms required reduction of pentavalent arsenic to trivalent before methylation could proceed, Vahter suggested that a similar mechanism might have been functioning in both the mice and rats. Evidence to support this hypothesis was presented in the author's observation that trivalent arsenic seemed more readily methylated than pentavalent.

Two recent studies, carefully designed for the specific purpose of addressing the oxidation/reduction issue for inorganic arsenic *in vivo*, have demonstrated that pentavalent arsenic (arsenate) undergoes reduction *in vivo* to the trivalent state in the rat (Rowland and Davies, 1982) as well as in mice and rabbits (Vahter and Envall, 1983). In both of these investigations, the issue of artifactual interconversion of these two valence states was carefully addressed.

In the rat (Rowland and Davies, 1982), administration of moderate doses of pentavalent arsenic led to rapid reduction to the trivalent form. In mice and rabbits, the rate of conversion was somewhat slower than in the rat, but still occurred at a significant rate. Trivalent arsenic was measured in arsenate-dosed mice plasma by 1 hour post-dosing.

In a summary report, Peoples (1983) reported, without experimental detail, that the feeding of sawdust containing chromated copper arsenate did not result in measurable urinary levels of trivalent arsenic. Since these animals were exposed to copper as well as chromium (VI), in addition to arsenate, it is difficult to differentiate any influence these agents may have had on the results. Peoples (1983) also noted that renal perfusion of dogs with a very dilute solution of arsenate did not lead to renal reduction of pentavalent to trivalent arsenic. In the absence of experimental details, it is not possible to determine what steps were taken to avoid artifactual conversion of trivalent back to pentavalent arsenic, if the lower valence form was indeed formed.

Studies by Lerman and Clarkson (1983) of penta- and trivalent arsenic in the rat are fully consistent with a reductive methylation process. Using *in vivo* and *in vitro* techniques, the authors showed that the isolated rat liver hepatocyte readily methylates trivalent arsenic, while virtually no conversion occurs with the pentavalent form, although it is known that pentavalent arsenic is also extensively methylated *in vivo*. These data suggest that arsenate must first be converted to arsenite elsewhere in the organism.

Two recent studies using human subjects and identical chemical speciation techniques for arsenic appear to offer conflicting data regarding pentavalent arsenic reduction.

Crecelius (1977) noted that ingestion, by a single subject, of a sample of well water tested as having 200 μg As only in the pentavalent form resulted in a urinary level of trivalent arsenic which was virtually the same as background concentration before taking the arsenic sample. Although urine was collected for approximately 3 days, a significant amount of the total dose was unaccounted for, making it difficult to draw any conclusions from the study.

Yamauchi and Yamamura (1979) fed a preparation of seaweed containing 86 percent pentavalent arsenic and 7 percent each of trivalent inorganic and dimethyl arsenic to 3 human subjects as one dose at a total dosing of 650, 680, and 760 μg total arsenic. By 48 hours, based on their method which was the Braman et al. (1977) procedure, 36 percent of the original dose appeared in urine, with 75 percent present as methylated forms and about 17 percent present as trivalent arsenic (6.3 percent of the total amount ingested). The authors calculated that the amount of trivalent inorganic arsenic was greater than could be ascribed to the small amount ingested originally and they stated that the fraction represented in vivo reduction of pentavalent arsenic. Since the mean intake was approximately 700 μg, the 7 percent present as trivalent inorganic arsenic amounted to 49 μg, of which 36 percent or 18 μg would have been expected to appear in urine in some form. Using 75 percent methylation, only approximately 4-5 μg would have been present as the trivalent form, when in fact a mean value of 43.4 was found (range: 36-51 μg). Since the Braman et al. (1977) method is a reasonably accurate speciating technique and there is little evidence that samples were handled in a way to promote artifactive formation of trivalent arsenic after excretion, the data cannot be readily questioned on this basis.

4.2.3 Chemical Stability of Trivalent and Pentavalent Inorganic Arsenic to Oxidation-Reduction

In aerated water, trivalent inorganic arsenic will undergo extensive oxidation to the pentavalent form, particularly when present at low levels (Feldman, 1979).

The pH of aqueous solutions appears to be the major factor in the relative stability of either valency form. Buchet et al. (1980) found that trivalent arsenic in solutions at pH of 7.0 or 9.6 were oxidized to the extent of 70-90 percent within one week, compared to 25 percent conversion at pH 4.8.

Vahter and Norin (1980) also noted rapid oxidation of As (III) in aqueous solution at room temperature, while storage at 4°C showed little conversion.

Pentavalent inorganic arsenic, on the other hand, is stable at neutral or alkaline pH but undergoes reduction with decreasing pH (Durrant and Durrant, 1966).

In studies directed to *in vivo* transformations of inorganic arsenic, urine levels are commonly used and some data exist regarding valency stability of arsenic in urine.

Buchet et al. (1980) found that trivalent inorganic arsenic in urine at pH 7.0 or less was relatively stable, with only 10 percent oxidation occurring by 7 days. At pH 9.5, 50 percent of oxidation occurred within one day.

4.3 DISTRIBUTION OF ARSENIC IN MAN AND ANIMALS

Blood is the main vehicle for transport of arsenicals from absorption sites to the tissues, with the hemokinetic character of arsenic being dependent on the animal species studied.

It is readily apparent from the literature that the rat constitutes an anomalous model for studies of the fate of inorganic arsenicals *in vivo* and this includes the clearance behavior of rat blood-borne arsenic (Hunter et al., 1942; Ducoff et al., 1948; Lanz et al., 1950; Ariyoshi and Ikeda, 1974;

Klaassen, 1974; Tsutsumi and Kato, 1975; Dutkiewicz, 1977). In the case of the rat, arsenic in blood is only slowly cleared following exposure, with about 80 percent of the total blood arsenic content localized in the erythrocyte. The half-times of blood clearance for inorganic arsenic in the rat (trivalent or pentavalent) is on the order of 60 to 90 days (Lanz et al., 1950; Ariyoshi and Ikeda, 1974).

Arsenic in the blood of other species--man (Ducoff et al., 1948; Mealey et al., 1959; Tam et al., 1979b), mice (Lanz et al., 1950; Crema, 1955), rabbit (Hunter et al., 1942; Ducoff, 1948; Klaassen, 1974), dog (Lanz et al., 1950; Hunter et al., 1942), and primates (Hunter et al., 1942; Klaassen, 1974)--whether given as the pentavalent form or as the trivalent form, is rapidly cleared. Normal blood arsenic values for individuals in the U.S. and Europe are in the range of 1-5 μg total arsenic/liter whole blood (Bergstromn and Wester, 1969; Damsgaard et al., 1973; Kagey et al., 1977; and Valentine et al., 1979). According to Kagey et al. (1977), cigarette smokers showed mean blood arsenic levels approximately 50 percent higher than nonsmokers. It is reasonable to assume that human background blood levels reflect mainly dietary arsenic, much of which would likely be in various organo-arsenical forms that are extensively absorbed and rapidly cleared.

Clearance of arsenic in dog and man was found to fit a three-compartment model by Charbonneau et al. (1978a) and Tam et al. (1979b) with half-times of 1, 5, and 35 hours, respectively. When contrasted with the work of Tam et al. (1978), which reported the time-dependent *in vivo* methylation of arsenic and excretion in dogs, the various components presumably relate to initial excretion of inorganic arsenic, followed by clearance of dimethyl arsenic.

The tissue partitioning of arsenic in man has been studied using both autopsy and dosing data. Kadowaki (1960) found (measurements in ppm, wet weight) that nails (0.89), hair (0.18), bone (0.07-0.12), teeth (0.08) and skin (0.06) generally housed the highest absolute amounts of arsenic, while heart, kidney, liver and lung contained somewhat lower levels (0.04-0.05). Brain tissue (0.03) had an arsenic level only slightly lower than other soft tissues. Liebscher and Smith (1968), analyzing tissue samples (ppm dry weight) from non-exposed sources in Scotland, observed lung to have the highest levels (0.09), with liver and kidney levels (0.03) not materially different from other soft tissue. Like the Kadowaki study, hair (0.46), nails (0.28) and skin (0.08) had the highest absolute values; however, bone and teeth (0.05) did not contain levels appreciably different from some other tissues (pectoral muscles, 0.06; ovary, 0.05; and pancreas, 0.05).

In addition to the autopsy studies by Kadowaki (1960) and Liebscher and Smith (1968), Larsen et al. (1979) have recently reported on a detailed study of the topographical distribution of arsenic in normal human brain tissue. Arsenic is distributed throughout all brain regions, with white matter showing higher levels than gray matter.

In looking at tissue distribution of arsenic in experimental animals, exposure of various species to either tri -or pentavalent arsenic leads to the initial accumulation of the element in liver, kidney, lung, spleen, aorta, and skin (Hunter et al., 1942; Ducoff et al., 1948; Lanz et al., 1950; Peoples, 1964; Ariyoshi and Ikeda, 1974; Cikrt and Bencko, 1974; Klaassen, 1974; Tsutsumi and Kato, 1975; Urakabo et al., 1975; Dutkiewicz, 1977; Sabbioni et al., 1979; Marafante et al., 1980). With the exception of the rat, a species in which metabolism of arsenic is only a very limited model for study of this element (vide supra), clearance from soft tissue is rather rapid except for the skin, where the high sulfhydryl group content may promote tight trivalent arsenical binding. As also seen with human tissue, arsenic is apparently lodged in brain of experimental animals exposed to arsenic, with slow clearance reported (Crema, 1955).

Recently, Vahter and Norin (1980) compared the dependency of dose and valency form of inorganic arsenic on tissue compartmentalization of arsenic in mice. Unlike most of the earlier reports, these workers used chemical speciation techniques (ion-exchange chromatography) to verify that their dosing media contained purely tri- or pentavalent arsenic. Using single oral dosing at 0.4 or 4.0 mg/kg levels and radioisotopic tri- or pentavalent arsenic, levels of arsenic in kidney, liver, bile, brain, skeleton, skin and blood were always greater (2-10 fold) in terms of percent total dose for the trivalent form than for the pentavalent form, and most pronounced at the higher dose. These workers ascribe much of this difference to relative methylating efficiency as a function of exposure level and valence form.

In a similar study using Golden Hamsters exposed to injected tri- or pentavalent radioisotopic arsenic, with care taken to assure valence purity, Ckirt et al. (1980) found that levels of arsenic in liver, kidneys, gut wall and bile were always greater, 2 to 25-fold, with trivalent arsenic exposure.

4.4 ARSENIC ACCUMULATION

The long-held view of arsenic as an element that accumulates in the body was mainly based on the behavior of arsenic in the rat, an animal model which in retrospect was the least helpful in understanding the fate of the toxicant _in vivo_ for other mammalian species and man.

Based on current arsenic elimination data for all mammalian species studied other than the rat (_vide supra_), one concludes that marked long-term accumulation of arsenic generally does not occur in physiologically vital components of the body. This is in contrast to, say, long-term lead accumulation in bone or cadmium accumulation in renal cortex. Autopsy tissue data for human subjects of different ages are not conclusive regarding possible long-term tissue accumulation. Kadowaki (1960) did observe higher mean levels of arsenic in skin and kidney samples of subjects approximately 50 years of age versus infant values, but dietary histories of the subjects were not available to allow for differentiation of increases in arsenic levels due to current versus past exposures for the older subjects. Deposition in hair is really excretory in nature, not accumulative.

Brune et al. (1980) have reported that lung tissue from retired smelter workers from the Rönnskär smelter in Sweden, on autopsy, had median values for arsenic which were about 8 times higher than that for a control group. Kidney and liver values, however, were not significantly different between smelter worker groups and controls. Arsenic accumulation in the lung of smelter workers even after several years of retirement and removal from workplace exposure (interval of 2-19 years) suggests that a very insoluble form of arsenic exists in smelter ambient air and is inhaled by these workers.

Lindh et al. (1980) measured the arsenic in autopsy femur samples of 7 occupationally exposed workers. The time between retirement and death ranged from 0-21 years. A control sampling from 5 autopsy cases was included. Using both neutron activation analysis and proton-induced X-ray emission (PIXIE) techniques, no clear evidence of significant arsenic accumulation in bone was seen. The scatter of levels was wide in the worker group, 0.006-0.21 ppm, with a median of 0.014 ppm, versus a range of 0.005-0.007 ppm in the control samples.

4.5 ARSENIC EXCRETION IN MAN AND ANIMALS

Renal clearance appears to be the major route of excretion of absorbed arsenic in man and animals, biliary transport of the element leading to enteric reabsorption with little carriage in feces.

Recent data for normal or background levels of arsenic in urine of human subjects in the U.S. reveal values of less than 20 μg/liter (20 ppb). If seafood has been consumed, such values can rise considerably to levels of 1,000 μg/liter or higher (Westöö and Rydälv, 1972; Pinto, 1976).

In a study designed to assess the utility of urine arsenic measurement in occupational exposure settings, Mappes (1977) reported excretion data for both single and multiple daily dosing for a human subject ingesting an arsenite solution. By 3 hours, renal excretion was maximal, with about one-quarter of the single dose appearing in the urine by day 1 post-exposure. With successive arsenite ingestion (0.8 mg As), daily urinary clearance after 5 days was about two-thirds of daily intake.

Buchet et al. (1981b), in their study of human subjects ingesting arsenite in amounts ranging from 125-1000 μg/day for 5 days, calculated a steady state occurring within 5 days. With steady state, 60 percent of daily intake is excreted daily. This figure is in good agreement with that of Mappes (1977) cited above.

Crecelius (1977) noted that following ingestion of arsenic in wine [50 μg As (III), 13 μg As (V)], approximately 80 percent of the dose was excreted within 61 hours. Oral ingestion of arsenic (V) in well water (200 μg), however, led to about 50 percent urinary excretion by 3 days post-ingestion. Mealey et al. (1959) measured urine arsenic in patients given trivalent arsenic by intravenous administration, with approximately 60 percent of the dose amount appearing in the urine within 24 hours. Hunter et al. (1942) noted that a group of human subjects given arsenic via parenteral administration exhibited considerable variance in the urinary clearance of arsenic, ranging from 30 to 80 percent after 4 to 5 days.

As might be predicted from the *in vivo* behavior of arsenicals in the rat, urinary excretion of arsenic in this species is very slow (due to erythrocyte retention) on the order of two to five percent of the arsenic intake by several days post-dosing (Coulson et al., 1935; Ariyoshi and Ikeda, 1974). Urakabo et al. (1975) calculated a half-time of 84 days for arsenic in the rat.

Slow clearance of arsenic from the rat gave rise to the widely-held assumption for many years that arsenic was one of the elements that accumulated in the body. Other species excrete arsenic rapidly. Mice, rabbits, swine, dogs, and monkeys clear the majority of injected trivalent arsenic within 24 hours, with excretion usually being ≥ 70 percent within that time period (Ducoff et al., 1948; Crema, 1955; Munro et al., 1974; Lakso and Peoples, 1975; Tam et al., 1978; Charbonneau et al., 1978a). Other studies also indicate rapid urinary clearance of arsenic given in the pentavalent form to species other than the rat (DuPont et al., 1942; Ginsberg and Lotspeich, 1963; Peoples, 1964; Lakso and Peoples, 1975).

Several studies have compared the relative rates of elimination of radiolabeled (^{74}As) trivalent and pentavalent inorganic arsenic (Vahter and Norin, 1980; Vahter, 1981) where precautions were taken to assure the purity of the respective dosing solutions as to valency state. Whole-body retention of trivalent inorganic arsenic in mice was 2-3 times greater than that of arsenate while retention times for both forms were dose-dependent, increasing with increasing exposure. In these studies, the animals received single oral doses of either form at levels of 0.04 to 4.0 mg As/kg. Differences in elimination rates appeared to relate to the relative degree of methylation of inorganic arsenic to the rapidly excreted methylated forms, chiefly dimethyl arsenic. Furthermore, differences in excretion rates were dose-dependent. At 0.04 mg As/kg, there was no difference between tri- and pentavalent arsenic elimination (Vahter, 1981).

Biliary transport of arsenic has been reported for a number of species. Bile-excreted arsenic is reabsorbed. Cikrt and Bencko (1974) noted that the rat had a higher biliary excretion rate for the trivalent than for the pentavalent form (approximately 10:1). Klaassen (1974) noted that the biliary excretion rate was much greater for the rat than for either the rabbit or the dog.

Cikrt et al. (1980) monitored biliary exretion in the Golden Hamster using penta- or trivalent inorganic arsenic. Significant differences in biliary excretion rates and cumulative excretion were seen between the two forms, being much greater for the trivalent form. However, fecal and urine arsenic content were greater with pentavalent arsenic administration. Biliary transport data for man are not available.

Deposition of arsenic in such organs as hair and nails can be considered an excretory mechanism for arsenic. Although hair analysis has had a long history in the chemical and forensic arsenic literature, for reasons of both analytical convenience and the possibility of establishing an exposure history from sectional analysis, many questions remain unanswered. The relationship between arsenic deposition in hair and various exposure parameters has not been well defined on a quantitative basis, nor are the physiological mechanisms well understood. The chemical nature of hair arsenic is also largely unknown.

5. ARSENIC TOXICOLOGY

The acute and chronic toxicity of arsenic will largely depend on the chemical form and physical state of the compound involved. Inorganic trivalent arsenic is generally regarded as being more acutely toxic than inorganic pentavalent arsenic, which in turn is more toxic than methylated forms of arsenic (NAS, 1977; Pershagen and Vahter, 1979; WHO, 1981). The previously noted (Chapter 3) "fish arsenic" is regarded as nontoxic. It is thus necessary to always specify, if possible, arsenic compounds when discussing effects and constructing dose-effect and dose-response relationships. In addition, factors like particle size and solubility must be taken into account. Trivalent compounds with low solubility, e.g., arsenic sulfide, will have low oral toxicity but may be retained in the lung (Pershagen et al., 1982).

The following chapter discusses the non-mutagenic/non-carcinogenic effects of exposure to inorganic arsenic. Because of the large volume of information available regarding the mutagenic and carcinogenic effects of inorganic arsenic, these topics have been discussed in the following chapters.

5.1 ACUTE TOXICITY OF ARSENIC IN MAN AND ANIMALS

In animal experiments the oral LD_{50} has been found to vary from 15 to 293 mg/kg body weight in rats and from 10-150 mg/kg in other animals (Dieke and Richter, 1946; Harrison et al., 1958). The lower values refer to experiments with soluble arsenic compounds. Franke and Moxon (1936) found that the LD_{75} 48 hours after i.p. administration was 4-5 mg As/kg body weight for sodium arsenite and 14-18 mg/kg for sodium arsenate.

Acute effects seen in animals after oral exposure are similar to effects seen in human beings and include gastroenteritis, diarrhea and cardiovascular effects (Nelson et al., 1971; Selby et al., 1977).

A large number of acute arsenic poisonings are described in the literature, but few data exist on actual doses and type of compound. Vallee et al. (1960) estimated the lethal dose to be on the order of 70 to 180 mg for arsenic trioxide. Acute symptoms are due to severe gastrointestinal damage, resulting in vomiting and diarrhea and general vascular injury which may lead to shock, coma or even death. Other acute symptoms are muscular cramps, facial edema, and cardiovascular reactions (Holland, 1904; Done and Peart, 1971).

Acute symptoms have also been seen after airborne exposure to high concentrations of arsenic trioxide, causing severe irritation of nasal mucosa, larynx, and bronchi (Holmqvist, 1951; Pinto and McGill, 1953).

Reversible effects on the hematopoietic and cardiovascular systems and peripheral nervous disturbances with slow recovery have also been noted (Ohta, 1970; Heyman et al., 1956; Jenkins, 1966; Nagamatsu and Igata, 1975: O'Shaughnessy and Kraft, 1976; Hamamoto, 1955; Chhuttani et al., 1967; LeQuesne and McLeod, 1977).

Over the years, a number of large-scale poisonings have occurred due to contamination of beer (Kelynack et al., 1900), soy sauce (Mizuta et al., 1956), dried infant milk (Tokanehara et al., 1956), and well water (Yoshikawa et al., 1960). These episodes mainly caused subacute and chronic symptoms and will be discussed in the following sections.

5.2 CHRONIC TOXICITY OF ARSENIC IN MAN AND ANIMALS

5.2.1 Neurotoxic Effects

Arsenic neurotoxicity, including both peripheral and central nervous system injury, has long been recognized as being associated with acute, subacute, and chronic exposures to relatively high levels of inorganic arsenic. These have been well characterized as to their major pathophysiological features, clinical course, sequelae and associated histopathology.

Reynolds (1901) provided one of the earliest detailed descriptions of arsenic-induced neurotoxic effects in his report on the clinical assessment of more than 500 patients who had consumed arsenic-contaminated beer.

Neurological involvement started with sensory changes, e.g., paresthesias, hyperesthesias, and neuralgias, accompanied by considerable muscle tenderness. Varying degrees of motor weakness, progressing from distal to proximal muscle groups, also occurred and culminated at times in paralysis of affected muscle groups or extremities. Certain indications of central nervous system (CNS) damage, e.g., loss of memory and general mental confusion, were also observed but were discounted by Reynolds (1901) as being less likely due to arsenic than chronic alcoholism or concurrent excessive selenium intake.

Peripheral nervous system (PNS) effects similar to those described by Reynolds (1901) have since been observed in numerous other cases of acute, subacute, and chronic arsenic exposures (Silver and Wainman, 1952; Mizuta et al., 1956; Heyman et al., 1956; Jenkins, 1966; Hara et al., 1968; Chhuttani et al., 1967; Ishinishi et al., 1973; Nakamura et al., 1973; Nagamatsu and Igata, 1975; O'Shaughnessy and Kraft, 1976; Frank, 1976; Garb and Hine, 1977; LeQuesne and McLeod, 1977) and are now recognized as classic clinical symptoms of arsenic poisoning. Such symptoms include peripheral sensory effects characterized by the appearance of numbness, tingling, or "pins and needles" sensations in the hands and feet, as well as decreases in touch, pain, and temperature sensations in a symmetrical distribution. These symptoms are often variously accompanied by burning sensations, sharp or shooting pains, and marked muscle tenderness in the extremities. Peripheral neuritis symptoms originate distally and, over the course of a few weeks, often progressively become more widespread in both lower and upper extremities, usually appearing first in the feet and later in the hands.

Collectively, the above components of the classical clinical syndrome associated with excessive arsenic exposure are highly indicative of progressive peripheral polyneuropathy, involving both sensory and motor nerves, and most intensively affecting long-axon neurons. In addition, biopsy and autopsy studies have provided histopathological evidence verifying peripheral nerve damage, especially Wallerian degeneration of long-axon myelinated nerve fibers in cases of human arsenic exposure where frank neurological signs and symptoms were manifested (Heyman et al., 1956; Jenkins, 1966; Chhuttani et al., 1967; Ohta, 1970; LeQuesne and McLeod, 1977).

The pattern of development of peripheral neuropathic effects is closely associated with the type of exposure. Acute exposure to a single high dose of arsenic can produce a fairly rapid onset of both motor dysfunctions and paresthesias. In one report, the onset was within 10 days (LeQuesne and McLeod, 1977) after exposure. Slow, incomplete recovery is usually seen in these cases.

Under more chronic occupational exposure conditions to lower levels of arsenic compounds, the development of neuropathy symptoms can be more gradual and insidious, and not only bilateral, but unilateral polyneuropathies without motor paralysis have been reported (Ishinishi et al., 1973; Nakamura et al., 1973). Again, the time course for recovery from the neuropathies, once induced, tends to be slow and on the order of years. Gradual onsets of peripheral neuropathies and slow recoveries have also been reported with subacute or chronic exposures to arsenic via ingestion of contaminated soy sauce (Mizuta et al., 1956) or anti-asthmatic herbal preparations containing arsenic trioxide or arsenic sulfide (Tay and Seah, 1975).

It is difficult to determine the levels of arsenic associated with the induction of peripheral neuropathies. For subacute or chronic poisoning situations, information has been provided in only a few studies by which effective exposure parameters can be estimated. Mizuta et al. (1956), for example, reported that peripheral neuropathies occurred in 20 percent of 220 patients of all age groups poisoned by ingestion of arsenic-contaminated soy sauce, with approximately 3 mg arsenic (likely as calcium arsenate) estimated to be ingested daily for 2-3 weeks, resulting in total effective doses up to approximately 60 mg. Also, Tay and Seah (1975) reported polyneuropathies in approximately 50 percent of 74 patients poisoned by daily ingestion of 3.3 or 10.3 mg/day of arsenic trioxide or arsenic sulfide in anti-asthmatic medicinal pills.

More subtle peripheral neurotoxic effects arising from chronic exposure to lower levels of arsenic in occupational or non-occupational groups are more difficult to establish, particularly as indexed by abnormal electromyographic or nerve conduction velocity findings. In one such study, Landau et al. (1977) reported relationships between length and intensity of occupational arsenic exposure (mainly to arsenic trioxide via inhalation) of smelter workers and alterations in peripheral nerve functioning. The manner in which the data were reported, however, precludes precise characterization of dose-effect/dose-response relationships.

In their study of arsenic in the drinking water of residents from Millard County, Utah, Southwick et al. (1981) conducted neurological examinations on all study participants 47 years of age and younger (see Section 7.1.2.2 for discussion of the total study population). Neurological examination revealed that conduction velocities for nerves studied (ulnar motor, median motor, ulnar sensory, median sensory, peroneal, sural) did not vary significantly with respect to age or community. In those individuals that exhibited below-normal conduction velocities, a slightly greater proportion was seen in exposed participants--12 percent of 67 controls versus 16 percent of 83 exposed. The sural nerve seemed most often to be affected even after adjusting by a correction factor of 1.8 m/s/degree in individuals who had nerve temperatures below 30° C. (Velocities below 37 m/s at 30° C or above were considered abnormal). Slowing of sural nerve conduction was reported equally in exposed and control participants. Nerve conduction velocities regressed against annual arsenic dose and the log of the dose showed no significant associations.

Similar difficulties have been encountered in attempts to characterize dose-effect/dose-response relationships for arsenic-induced peripheral nerve functional deficits (as demonstrated by electromyographic recording techniques) in studies of two other populations chronically exposed to arsenic: (1) a population of Yellowknife Canadian Indians exposed via occupational contact with arsenic in a gold mining and smelting facility, or, in the case of the families of such workers, via arsenic emissions from the facility into the ambient environment (Canadian Public Health Assoc., 1978); and (2) a Nova Scotia population exposed, via geological arsenic contamination of wells, to levels >0.05 ppm arsenic in drinking water (Hindmarsh et al., 1977).

Several of the clinical reports discussed above not only document peripheral nerve damage but also contain descriptions of arsenic-induced central nervous system (CNS) disturbances or encephalopathy effects ranging in severity from memory losses and general mental confusion to convulsions, stupor, coma and even death (Heyman et al., 1956; Jenkins, 1966: Frank, 1976; Nagamatsu and Igata, 1975; O'Shaughnessy and Kraft, 1976; Garb and Hine, 1977). The onset and courses of such CNS effects have not been well defined, but they appear to closely parallel the development of peripheral neuropathy effects. Cases of prolonged encephalopathy indexed by electroencephalogram (EEG) recordings of abnormal brain wave patterns up to a year after cessation of exposure have been reported (Freeman and Couch, 1978; Bental et al., 1961). Such effects appear to be a much less constant feature of arsenic-induced neurotoxic effects in adults than are peripheral neuropathies.

Certain studies suggest, in contrast, that children may be more susceptible to arsenic-induced CNS damage. For example, severe CNS deficits have been observed in children exposed for several months as babies to arsenic-contaminated powdered milk formulas in Morinaga, Japan (Hamamoto, 1955; Okamura et al., 1956; Yamashita et al., 1972; Masahiki and Hideyasu, 1973; Japanese Pediatric Society, 1973). Follow-up studies on the children exposed to arsenic as infants have revealed: (1) increased incidence of severe hearing loss (>30 dB) in 18 percent of 415 children examined compared to less than 1 percent incidence of hearing loss in corresponding age-group children; (2) increased incidence of abnormal electroencephalographic (EEG) brain wave patterns in 14 percent of the exposed children, more than double the expected rate for comparable normal pediatric populations; and (3) observations of increased incidences of persisting mental retardation, epilepsy, and other indications of severe brain damage.

In another study (Bencko and Syman, 1977), hearing losses in children were reported to be associated with arsenic exposure derived from emissions from a nearby coal-fired power plant combusting high-arsenic-content coal. Both air and bone conduction hearing losses were observed, suggesting inner ear damage. Failure to find analogous hearing losses in children exposed to atmospheric arsenic emitted from a copper smelter in the United States (Milham, 1977) has raised questions regarding arsenic-induced damage to the inner ear in children.

Very few animal toxicology studies have focused on investigation of neurotoxic effects of arsenic on the CNS. Rozenshtein (1970), for example, reported evidence of CNS functional deficits, as indexed by altered conditioned reflexes, as well as histopathologic evidence of CNS structural damage, e.g,. pericellular edema and neuronal cytolysis in the brain, in rats exposed for three months to an arsenic trioxide aerosol resulting in an arsenic concentration of 46 $\mu g/m^3$. Similar but less severe effects were also obtained with exposure of other rats to a 3.7 $\mu g\ As/m^3$ aerosol. CNS deficits, indexed by impaired avoidance conditioning in the absence of demonstrable histopathologic changes in brain tissue, were also reported (Osato, 1977) for suckling rats administered 2 or 10 mg arsenic trioxide via stomach intubation over a 40-day period.

5.2.2 Cardiovascular Effects

A specific cardiovascular effect is "Blackfoot" disease, a peripheral vascular disease leading to gangrene of the toes, feet, legs and fingers. This disease has been reported in an area of Taiwan, where exposure to inorganic arsenic via drinking water is known to occur. In their skin cancer study of the Taiwanese population (see Section 7.1.2.2 for complete discussion of the total study population), Tseng and co-workers (1968, 1977) also investigated the occurrence of Blackfoot disease. The total prevalence of Blackfoot disease was lower than the prevalence of skin cancer, but higher in the younger age groups (Table 5-1). The overall prevalence rate for Blackfoot disease was 0.9-1.2 percent for males and 0.7 percent for females after age 39.

A group of 1108 patients with Blackfoot disease from the endemic area was identified during 1958 to 1975 (Tseng, 1977). There were 669 males and 439 females. Patients were included in the series if they had: 1) objective signs of ischemia, and 2) subjective symptoms of ischemia. Follow-up was attempted using a variety of methods to trace the subjects. At the end of follow-up, 528 patients had died, a fatality rate of 47.7 percent.

A history of typical ischemic symptoms such as numbness was used to estimate the date of onset of Blackfoot disease. Duration of intake of arsenic

TABLE 5-1. PREVALENCE OF BLACKFOOT DISEASE (per 1000) BY AGE AND ARSENIC EXPOSURE (ppm)

Arsenic content of drinking water (ppm)	Age 20-39	40-59	>60
<0.3	4.5	10.5	20.3
0.3 - 0.6	13.2	32.0	32.2
>0.6	14.2	46.9	61.4

Source: Adapted from Tseng, 1977.

water at the time of onset represents the period of time between first use of such intake and the time of onset of the disease. Duration of intake of arsenical water represents the duration of time during which the patient started drinking artesian well water up to the time of survey or up to the time of change of source of drinking water. For native patients, the duration is estimated to be equivalent to their ages, but for patients who came from areas without artesian wells the duration was counted as starting from the year of arrival.

A classification system of percentage of permanent disability was created and each patient was assigned to the appropriate category.

It was determined (determination not specified) that all patients with Blackfoot disease had consumed artesian well water before the onset of disease, and none of the residents of the endemic area who had consumed surface water or water from shallow wells developed Blackfoot disease.

In some parts of the arsenic endemic area, a new source of drinking water was provided in 1956. The incidence of Blackfoot disease was therefore examined in year of onset periods (1955 and before, 1956-1965, and 1966-1975). There were no cases of Blackfoot disease among area residents who were born after the tap water supply was instituted in 1956. Furthermore, as the duration of intake of arsenical water increased (across all 3 exposure levels) the incidence of Blackfoot disease increased. In addition, the degree of permanent disability of patients was significantly correlated with duration of intake of arsenical water at time of onset of the disease.

There are also data indicating that other substances may be involved in the etiology of Blackfoot disease. Lu et al. (1977, 1978) found fluorescent compounds in water samples from an area with endemic Blackfoot disease. Of the compounds discovered, tentative identifications of D-lysergic acid and/or ergometrine, ergotamine and calciferol have been made (Irgolic, 1982). These substances may have been at least contributing factors to this disease, since some of them, or similar compounds, are known vasoactive agents.

Vascular changes were also noted among persons living in Antofagasta, Chile (see Section 7.1.2.2 for discussion of study groups). Raynauds syndrome and acrocyanosis were reported to occur in 30 and 27 percent, respectively, in a group of 100 persons studied by Borgono and Greiber (1972).

Raynauds syndrome has also been reported to occur among German vintners. Butzengeiger (1940) studied 180 persons and found that 22.8 percent had evidence of vascular disorders of the extremities. Butzengeiger (1949) studied 192 vinegrowers and found that 28.7 percent had ECG changes. There was, however, no control group.

In epidemiological studies of smelters (see Section 7.1.2.1 for complete discussion of studies), peripheral vascular disease has generally not been found. However, mortality studies indicate that there might be some cardiovascular effects, although study results have been conflicting. Thus, Axelson et al. (1978) found a higher mortality in cardiovascular disease, but analysis of the data indicate that if such an effect occurred it probably needed higher exposure levels than those causing lung cancer. A study by Ott et al. (1974) indicated, on the other hand, that the arsenic-exposed workers had less mortality than expected.

In studies on workers at the Anaconda Copper Smelter in Montana, both Lee and Fraumeni (1969) and Lee-Feldstein (1983) reported that cardiovascular mortality was significantly elevated in the workers--SMR=118 and SMR=129, respectively ($p < 0.01$)--but not related to duration of arsenic exposure. In a study cohort of these same workers, Higgins et al. (1982) reported that cardiovascular mortality increased with increasing ceiling arsenic exposure among smokers, but not among nonsmokers--at 500-4999 ug/m^3, SMR=165 for smokers ($p < 0.01$), SMR=100 for nonsmokers; at >5000 ug/m^3, SMR=182 for smokers ($p < 0.01$), SMR=166 for nonsmokers. In contrast, Lubin et al. (1981) did not find an excess in cardiovascular disease mortality (SMR=108) in their study cohort of workers from the same smelter. The cohorts in all four of these studies were slightly different.

The conflicting findings of these reports suggest that the relationship between arsenic exposure and cardiovascular disease is quite complex and in need of additional research.

5.2.3 Teratogenesis and Developmental Effects

5.2.3.1 Animal Studies--In a recent review of the toxicological effects of prenatal exposure to arsenic, Hood (1983) discussed the number of variables that influence effects on offspring of maternal arsenic exposure during pregnancy. Included in the variables are the forms of arsenic administered, species and individual differences in susceptibility, dose level and exposure route, peak level attained in the conceptus, maternal metabolism and excretion, and timing of exposure during gestation; the length of the list being indicative of the complex nature of arsenic toxicity induced during prenatal exposure.

Teratogenic effects of arsenic compounds, at least at relatively high exposure levels, have been demonstrated in a number of animal species. Studies by Ridgeway and Karnofsky (1952), for example, demonstrated no gross abnormalities in chick embryos following injection of sodium arsenate into embryonate eggs at 0.20 mg As/egg on day 4 of gestation. Retardation in body weight gain and feather growth, as well as certain other abnormalities, were, however, observed in chickens hatched from arsenic-treated eggs.

Later experimental studies on mammalian species have demonstrated teratogenic effects of arsenic in hamsters, rats and mice.

Ferm and Carpenter (1968) produced malformations in 15-day hamster fetuses via intravenous (I.V.) injections of sodium arsenate into pregnant dams on day 8 of gestation at dose levels of 15, 17.5 or 20 mg/kg body weight. The variety of malformations obtained with prenatal arsenic exposure of hamsters--including exencephaly, encephaloceles, skeletal defects and genitourinary system defects and the effects of other related experimental manipulations, e.g., protective effects of co-treatment with selenium--were reviewed in a later report by Ferm (1977).

Willhite (1981) injected arsenate (20 mg/kg) or arsenite (2-10 mg/kg) into pregnant golden hamsters at day 8 of gestation and observed axial skeletal disorders in offspring--cranioschisis aperta with exencephaly and cranioschisis occulta--while 10 hours after dosing, embryos showed a delay in neural fold elevation and neural tube closure with arsenate exposure.

Teratogenic and embryotoxic effects of prenatal arsenic exposure of mice have also been reported (Hood and Bishop, 1972; Hood and Pike, 1972; Hood et al., 1977). Increased fetal resorption, decreased fetal weights and various malformations (such as exencephaly, micrognathia, agnathia, exophthalmos, anophthalmia, hydroencephaly, cleft lip, ectrodactyly, micromelia, fused vertebrae and forked ribs) were observed following single intraperitoneal (I.P.)

injections of sodium arsenate (45 mg As/kg body weight) in Swiss-Webster mice (the single injection occurring on one day between days 6 and 12 of gestation) (Hood and Bishop, 1972). Co-treatment with 2,3-dimercaptopropanol (BAL) reduced arsenic-associated malformations (Hood and Pike, 1972; Hood et al., 1977).

Many similar embryotoxic and fetal teratogenic effects, along with other effects (renal and gonadal agenesis), have also been observed following I.P. injections of sodium arsenate in pregnant rats (Beaudoin, 1974; Burk and Beaudoin, 1977), again at relatively high dose levels (e.g., 30 mg/kg body weight).

Compared to pentavalent arsenic, trivalent arsenic as the arsenite salt appears to be proportionally more embryotoxic. For example, the fetotoxic effects seen by Baxley et al. (1981) in mice exposed to 40-45 mg/kg As as arsenite were comparable to effects observed by Hood et al. (1978), in which a much higher level of arsenate (120 mg/kg As) was administered.

The above studies are suggestive of significant effects on reproduction and development of mammalian species being induced by prenatal arsenic exposure. However, only very minimal effects, or none at all on fetal development, have been observed in studies on chronic oral exposure of pregnant rat or mice dams to relatively low levels of arsenic via drinking water (Schroeder and Mitchener, 1971). Nadeyenko et al. (1978) reported that intubation of rats with arsenic solution at a dose level of 0.0025 mg/kg for a period of 7 months, including pregnancy, produced no significant embryotoxic effects and only very infrequent slight expansion of ventricles of the cerebrum, renal pelvises and urinary bladder. Also, Hood et al. (1977) reported that very high single oral doses of arsenate solutions (120 mg/kg) to pregnant mice were necessary to cause prenatal fetal toxicity, while multiple doses of 60 mg/kg on 3 days had little effect. This report suggests that the higher doses required with oral versus parenteral exposure relate to the more rapid methylation to less toxic forms of oral arsenic owing to its initial passage through the liver, where methylation may occur. Similar results were obtained with mice in a later study (Hood et al., 1978).

Given the presence of biomethylation in mammalian species exposed to arsenic, the question arises as to whether fetotoxicity resides only in the untransformed inorganic arsenic or is due to some of the metabolites formed _in vivo_. Hood et al. (1982a) have demonstrated that methylated metabolites cross the placental barrier in mice given arsenite and that these methylated forms are actually the major form of arsenic in fetuses by 12 hours post-dosing.

Recently, three groups of investigators have reported their results on the comparative teratogenicity of methylated arsenicals, administered as such, to different species.

Willhite (1981) found no teratogenic response when pregnant golden hamsters were given methylated arsenic at single doses up to a level of 100 mg As/kg. Rogers et al. (1981) administered dimethylarsenic to pregnant CD rats and CD-1 mice on days 7-16 of gestation via oral gavage. Dosing was from 7.5 to 60 mg/kg/day for rats and 200 to 600 mg/kg/day for mice. Fetal and maternal toxicity was observed in both species. A teratogenic response in mice was confined to cleft palate in the 400- and 600-mg regimens. In the rat, there was a dose-dependent incidence of irregular palatine rugae.

Hood et al. (1982b) exposed pregnant golden hamsters to either dimethyl- or monomethyl arsenic at dosages of either 900 or 1000 mg/kg of the former agent and 500 mg/kg of the latter. The levels of dimethylarsenic administered were seen to be acutely toxic to dams and were associated with significant resorption rates and malformations in survivors. In the case of the monomethyl arsenic, the overall teratogenic potency was observed to be lower.

Harrison et al. (1980) exposed CD-1 mice to monomethyl arsenic by injection (1500 mg/kg salt, i.p.) on one of gestation days 8, 9, or 10. Treatment at days 9 and 10 of gestation resulted in increased resorption and mortality rates of 37 and 43 percent, respectively, while the day 8 group showed no such effects. Skeletal malformations were also seen at the 9 and 10 gestation day exposures. While details for dimethylarsenic were not provided, it was noted that such treatment also resulted in fetotoxicity and teratogenicity, with exencephaly the primary gross malformation observed.

The available data, in summary, indicate that while the methylated arsenicals are less teratogenic and fetotoxic on an equivalent dose basis, the nature of the effects is comparable. There also appears to be a species sensitivity difference for arsenical teratogenicity and fetotoxicity, the rat being more sensitive than mice or golden hamsters.

Animal studies on the effects of early postnatal administration of arsenic compounds on growth and development have generally failed to yield any significant positive results at sublethal arsenic dose levels. Tamura (1978), for example, reported no effects on the growth and development of postnatal rats fed arsenic trioxide in their diet from the 7th to 21st day following birth at a dose level of 1.5 mg/kg/day, in comparison to a 50 percent mortality rate at a 15.0 mg/kg body weight dose. Similarly, Ferslew and Edds (1979) observed no significant effects on growth or development of young swine fed arsanilic acid at 0.01 percent concentration for 31 days commencing at 3 weeks of age, despite observations of significant alterations by organo-arsenic of white blood cell counts, serum alkaline phosphatase activity, and other blood chemistry parameters and significant increases of arsenic in various soft tissues and urine. In another study (Heywood and Sortwell, 1979), both 3.75 and 7.5 mg/kg/day doses of an arsenic compound fed to adolescent or infant rhesus monkeys resulted in mortality in most of the exposed animals, but no notable effects in growth or development of animals surviving the dosing period. Unfortunately, the lack of experimental data in the above studies on the effects of exposure across a range of dose levels, besides the ones employed, greatly limits their utility in terms of determining possible dose-effect or dose-response relationships between postnatal arsenic exposure and induced effects on growth and development.

It should be noted that, whereas some of the above animal studies provide highly suggestive evidence for arsenic effects on reproduction and development, at least at high exposure levels, one cannot confidently extrapolate the results to estimate the probability of occurrence of similar or analogous effects in man. Still, some suggestions of possible arsenic effects on human reproduction and development have been derived from certain epidemiology studies discussed below.

5.2.3.2 Human Studies--Available data are mainly from studies in Sweden on male and female smelter workers. These studies were not designed specifically to study effects of arsenic but rather to study the effects, in general, of the smelter work. While data from these studies suggest a low-level effect of smelter pollutants on neighboring (proximate) populations, the diverse agents involved preclude making conclusive statements about the specific effects of arsenic.

Congenital malformations were found to occur at about the same rate, 3.0 percent among women employed at the Rönnskär smelter, as among women living in six areas belonging to the hospital serving the Rönnskär area (Nordström et al., 1978c). However, mean birthweights were lower among offspring to female Rönnskär workers (Nordström et al., 1978a, 1978b).

The number of spontaneous abortions was also increased among women working in this smelter; the highest rate (17 percent) was found among women employed during pregnancy or who had been employed prior to pregnancy and lived close to the smelter. Women working in close connection with smelting processes had a rate of 28 percent of spontaneous abortion compared with other female employees. When both parents were employed, the abortion rate was 19.4 percent compared to 13.5 percent when the father was not employed.

Studies on spontaneous abortion among women living in the vicinity of the smelter were also conducted (Nordström et al., 1978d). In four areas, the rate varied between 7.6 and 11.0 percent. The highest rate occurred in the area closest to the smelter, but many women employed at the smelter live in that area.

5.2.4 Hematological Effects

The hematopoietic system in man has been shown to be affected by arsenic exposure in cases of acute, subacute or chronic intake, taking the form of anemia, leukopenia, granulocytopenia and eosinophilia. Such effects appear to be reversible, the system recovering in a matter of weeks after exposure ends.

Hamamoto (1955), in his report of infant poisoning by arsenic-contaminated milk in Japan, described marked anemia and leukopenia with lymphocytosis in most of a group of 59 infant patients. Erythropoiesis was disturbed in bone marrow of 19 children studied. Within a month, the hematology appeared to show normal values. Similarly, Mizuta et al. (1956) found blood disturbances, including anemia with leukopenia and lymphocytosis, in subjects surveyed for ingestion of soy sauce contaminated with arsenic.

Two recent studies dealing with acute and subacute arsenic poisoning (Feussner et al., 1979; Lerman et al., 1980) have demonstrated arsenic-induced megaloblastic anemia. Feussner et al. (1979) demonstrated that arsenic was present in bone marrow by using electron-probe microanalysis.

Chronic exposure to arsenic occasions hematological effects which resemble those seen with subacute exposure. Terada (1960), in his survey of patients exposed to arsenic in well water contaminated by industrial activity in Niigata, Japan, saw anemia as a common feature, with the anemia being either normochromic (50 percent) or hyperchromic (30 percent). In their study on individuals living in communities in Utah where arsenic was present in drinking water, Southwick et al. (1981) also reported the presence of anemia in certain individuals; however, anemia was not significantly more prevalent in exposed populations. Anemia has been reported in subjects exposed to arsenic occupationally or in medicinals (Kyle and Pease, 1965; Westhoff et al., 1975).

In animals, decreased hemoglobin production has been seen in rats fed both arsenate (Mahaffey and Fowler, 1977) and arsenite (Byron et al., 1967) and in cats given either form orally (Massmann and Opitz, 1954). The study of Woods and Fowler (1977), in which arsenate was given orally to rats and mice at 20, 40 or 85 ppm in drinking water, showed that the sites of disturbance of the heme biosynthetic pathway by arsenate mainly involved depression of ALA-synthetase and heme-synthetase activity, with elevation in the urinary levels of chiefly uroporphyrin and also coproporphyrin. The increase in uroporphyrin appears to be specific to arsenic.

5.2.5 Hepatic Effects

In the reports of Hamamoto (1955) and Mizuta et al. (1956), dealing with arsenic contamination in infant milk and soy sauce, respectively, swollen livers appeared to be a common clinical feature. Of the children exposed to milk arsenic, all subjects presented with this feature (61/61). In the autopsies of fatal outcome cases, hemorrhagic necrosis and fatty degeneration of the liver were seen. In the survivors, liver function tests were not very revealing, being within normal limits in most cases. In both of the above episodes, liver size returned to normal after exposure ceased.

Over the years, chronic intake of arsenic has been reported to be associated with hepatic damage in the form of portal hypertension, malignant liver disease and cirrhosis. In the Manchester beer poisoning episode, Reynolds (1901) noted widespread liver disease in drinkers of such beer, with the extent of the disease being related to the amount of arsenic in the beverage.

The incidence of hepatic cirrhosis among German vintners has been reviewed by Luchtrath (1972), who noted that the frequency of liver cirrhosis decreased with the banning of arsenical pesticides. The use of arsenic as an anti-syphilitic is also known to be associated with hepatic cirrhosis (Baldridge, 1934).

Non-cirrhotic portal hypertension has been infrequently noted in the literature (Morris et al., 1974; Szuler et al., 1979). Szuler described one case report of presinusoidal portal hypertension in a patient taking an arsenic antiasthmatic for 55 years. Cirrhosis was absent and liver function was not disturbed.

In the smelter study by Axelson et al. (1978) there was a tendency towards an increased mortality in liver cirrhosis among arsenic-exposed workers.

5.2.6 Renal Effects

Functional and/or biochemical impairment of the human renal system is mainly seen in acute poisoning by arsine, AsH_3 (Uldall et al., 1970; Fowler, 1977). Oligouria and anuria progressing to renal failure are common acute responses. Persistent renal sequelae in subjects surviving the acute stage of arsine poisoning include chronic renal insufficiency and hypertension. Histologically, the injury is mainly tubular and interstitial.

Acute or chronic renal effects associated with arsenate or arsenite exposure are less well characterized. Gerhardt et al. (1978) have recently described a case of acute arsenic poisoning from contaminated illicit liquor.

Acute renal bilateral cortical necrosis was diagnosed in the patient who survived the acute stage to eventually develop reduced renal size and cortical calcification.

In the clinical survey of Hamamoto (1955) of Japanese infants poisoned with arsenic-contaminated milk, possible renal injury was diagnosed by the presence of hematuria, leukocyturia and glycosuria (23.7 percent, 42.3 percent and 13.5 percent, respectively). Reversibility of these indices was apparent, with only three percent of the patients still showing pathological changes after one month. Terada (1960) also noted proteinuria in cases of arsenic poisoning in Niigata, Japan, arising from ground water contamination from an arsenic plant.

5.2.7 Respiratory Effects Other Than Cancer

Nasal septum perforation is a rapid tissue response in workers encountering high airborne arsenic levels (NAS, 1977; Hine et al., 1977; Lundgren, 1954).

In the detailed smelter worker study of Lundgren (1954), 1,276 subjects in a worksite with levels up to 7 mg As/m^3 showed two types of respiratory disturbances relating to the nature of the processing and presumably the form of the arsenic. Refined arsenic handling was associated with septal perforation and rhinopharyngolaryngitis, while workers in the roaster, furnace, and connector areas showed tracheobronchitis and signs of pulmonary insufficiency. In the latter group, exposure was mixed, including both arsenic and sulfur dioxide. A recent report has suggested, furthermore, that workplace arsenic may also be present as the sulfide (Smith et al., 1976).

Chilean children exposed to arsenic in drinking water (Borgono et al., 1977) showed a chronic cough and bronchitic history.

5.2.8 Immunosuppressant Effects

The role of inorganic arsenic as an immunosuppressant in man is mainly inferred from indirect data accumulated over the years (Arsenic. NAS, 1977).

First, the therapeutic utility of arsenicals, such as arsenite-based Fowler's solution in the treatment of steroid-responding disorders and as a lymphocytostatic agent, suggests action as an immunosuppressant.

Secondly, certain manifestations indicative of likely immune system disorders have been observed with arsenic exposure, (e.g., the occurrence of herpes simplex and chronic pulmonary infections) and suggest a role of arsenic as an immunosuppressant (Arsenic. NAS, 1977). Histories of chronic cough and bronchitis in Chilean children exposed to arsenic in drinking water (Borgono et al., 1977) would tend to support such a role.

5.3 FACTORS AFFECTING ARSENIC TOXICITY

The most widely recognized and studied arsenic interactive behavior is with selenium, and much of the early data has been reviewed by Levander (1977).

The known antagonistic relationship of arsenic and selenium in a number of animal species was first described by Moxon and co-workers (Moxon and Dubois, 1939; Dubois et al., 1940), who also demonstrated the utility of dietary arsenic supplementation in protecting livestock from the toxic levels of selenium in certain fodders. Both penta- and trivalent arsenic are equally effective in protecting against selenium toxicity, and do so regardless of the chemical form by which selenium exposure occurs.

An understanding of the *in vivo* mechanisms by which arsenic imparts a protective effect on selenium toxicity has only recently been gained.

The ability of arsenic to retard the formation of volatile selenium in the form of dimethyl selenide (Kanstra and Bonhorst, 1953) apparently involves inhibition of microsomal methyl transferase, an enzyme sensitive to arsenite (Ganther and Hsieh, 1974). An overall protective effect still exists, however, since arsenic promotes the biliary excretion of selenium (Levander and Baumann, 1966; Ganther and Baumann, 1962). According to Levander (1977), enhanced biliary clearance of selenium in the presence of arsenic probably involves an excretory conjugate of both, since selenium likewise enhances the biliary excretion of arsenic.

Rössner et al. (1977) showed a protective effect for arsenite against the cytotoxicity of selenite, using suspension cultures of mouse fibroblasts exposed to these agents at 10^{-5} to 10^{-11} M. Interestingly, selenite, in turn, had only a low protective effect against arsenite cytotoxicity.

Few data exist for interactive relationships between arsenic and other elements. In one of the few pertinent studies bearing on this issue, the effects of concomitant oral exposure to cadmium, lead and arsenic versus single-agent exposure effects have been reported by Mahaffey and Fowler (1977). Cadmium and arsenic together retarded weight gain in young adult rats to a greater extent than either element alone.

6. ARSENIC MUTAGENICITY

Various inorganic compounds of arsenic have been tested for mutagenicity in a variety of test systems ranging in complexity from bacteria to peripheral lymphocytes of exposed human beings. Information on these tests has been reviewed herein and elsewhere (Flessel, 1978; NAS, 1977; Leonard and Lauwerys, 1980; WHO, 1981). The following chapter is divided into five major sections discussing studies indicative of mutagenic damage, as well as interactions of arsenic with other mutagens and possible mechanisms of mutagenic action.

6.1 GENE MUTATION STUDIES

6.1.1 Prokaryotic Test Systems (Bacteria)

Arsenic does not appear to induce gene mutations in bacteria (Table 6-1). Sodium arsenite ($NaAsO_2$) was tested for its ability to induce reverse mutations in *E. coli* (Nishioka, 1975). Three bacterial strains auxotrophic for tryptophan were tested for growth in the absence of tryptophan after arsenic treatment. Nishioka reported that arsenic induced mutations in wild type cells and in cells deficient in excision repair ($uvrA^-$) but not in cells deficient in recombinational repair ($recA^-$). This suggested that the rec function is necessary for arsenic-induced mutagenesis in *E. coli*.

The observations by Nishioka were not reproducible in subsequent studies by Rossman et al. (1980). In these latter studies, the authors examined sodium arsenite-induced mutagenesis in five strains of *E. coli*, all auxotrophic for tryptophan and carrying different mutations for DNA repair capacities. In addition, Rossman et al. used three protocols for quantifying the presence of mutant cells: the spot test, the plate test and the fluctuation test. Arsenite did not increase the mutation frequency in any *E. coli* strain using any of the

TABLE 6-1. SUMMARY OF MUTAGENICITY OF ARSENIC: GENE MUTATIONS

Reference	Test system	Chemical Information	Results	Comments
Nishioka, 1975	Reverse mutation in _E_. _coli_ Strains: WP2 (trp^-) WP2 uvrA ($uvrA^-$, trp^-) CM571 ($recA^-$, trp^-)	$NaAsO_2$ 8.0×10^{-4} M in WP2 1.6×10^{-4} M in WP2 uvrA 1.6×10^{-4} M in CM571 Source: Nakarai Chemical LTD (reagent grade) Solvent: H_2O	$NaAsO_2$ positive for mutations in WP2 and WP2 uvrA but not in CM571. Increases were approximately 10x background.	1. Doses of arsenite were selected to kill 60-70% of cells. 2. Lack of mutagenesis in CM571 suggests that a rec function is required for mutagenesis in _E_. _coli_. 3. Rossman et al. (1981) were unable to reproduce these mutation studies. They claimed (a) that the induced mutation frequencies observed by Nishioka were within the normal range of background, (b) that they observed a 31-fol difference in cytotoxicity studies compared to Nishioka, and (c) that Nishioka used an incorrect method for determining mutation frequency.

TABLE 6-1. (continued)

Reference	Test system	Chemical Information	Results	Comments
Rossman et al., 1980	Induction of reverse mutations in E. coli at the trp locus. Assays / Strains Spot test: WP2 (trp^-) Plate test: $WP2_S$ (trp^-, uvrA-) Fluctuation test: WP6 (trp^-, $polA1^-$) WP10 (trp^-, $recA1^-$) $WP44_S$-NF (trp^-, $uvrA^-$, tif-1/sti^-). tif-1 mutation confers a thermally inducible SOS repair system resulting in normal mutation at 30°C and a 20-fold increase in mutation at 42°C.	$NaAsO_2$: 50 µl of 0.1 M in spot tests with $WP44_S$-NF (30°C), WP2, WP6, and WP10. $NaAsO_2$: 10 µl of 0.01 M in spot test $WP44_S$-NF, 10 µl of 0.1 M in spot test $WP44_S$-NF. $NaAsO_2$: 25 mM in plate tests WP2. $NaAsO_2$: 0.4, 1, 2, mM in fluctuation test WP2.	Arsenic did not increase the mutation frequency in any of the assays. Arsenic showed a dose-dependent decrease in mutations in $WP44_S$-NF at 42°C.	1. Results indicate that arsenic may inhibit SOS repair.
Lofroth and Ames, 1978 (abstract only)	Salmonella microsome assay	Arsenite and arsenate	Negative	1. No data available since information comes from an abstract.

TABLE 6-1. (continued)

Reference	Test system	Chemical Information	Results	Comments
Singh, 1983	Gene conversion at the trp locus and reverse mutation at the ilv locus in S. cerevisiae.	$NaAsO_2$ (0.1 M added to well, volume not indicated) Solvent: H_2O	Negative for conversion. Weak positive for mutation.	1. Study lacking sufficient experimental details.
Strain D7 = $\frac{a\ ade\ 2\text{-}40}{\alpha\ ade\ 2\text{-}119}\ \frac{trp\text{-}5\text{-}12}{trp\ 5\text{-}27}\ \frac{ilv\ 1\text{-}92}{ilv\ 1\text{-}92}$				
Amacher and Paillet, 1980	Forward gene mutation at the thymidine kinase (TK) locus in mouse L5178Y cells.	Na_2HAsO_4 Doses tested: (0.83 x 10^{-4}, 1.1 x 10^{-4}, 1.47 x 10^{-4}, 1.96 x 10^{-4}, 2.26 x 10^{-4} M) Source: Fisher Solvent: Normal saline	Negative	1. Exposed 3 hrs at 37°C. 2. Survival ranged from 66% at the low dose to 35% at the high dose. 3. Other metals such as cadmium, nickel, and trans-platinum were positive.
Rossman et al., 1980	Induction of forward mutations in Chinese hamster cells *in vitro* at HGPRT locus and the sodium-potassium ATPase locus.	$NaAsO_2$ (0.5 x 10^{-6} M, 2 days) ouabain resistance (5.0 x 10^{-6} M, 1 hour) ouabain resistance (20 x 10^{-6} M, 1.5 hours) thioguanine resistance (100 x 10^{-6} M, 25 min) thioguanine resistance	No increased mutation	1. UVC and MNNG used as positive controls. 2. Doses of arsenite used varied from slightly inhibitory to cell growth to fairly toxic.

TABLE 6-1. (continued)

Reference	Test system	Chemical Information	Results	Comments
Oberly et al., 1982	Forward gene mutation at the thymidine kinase (TK) locus in mouse L5178Y cells.	$NaAsO_2$ (3.8 x 10^{-6}, 7.7 x 10^{-6}, 11.5 x 10^{-6}, 15.4 x 10^{-6}, 19.2 x 10^{-6} M) Na_2HAsO_4 (7.5 x 10^{-5}, 9.7 x 10^{-5}, 11.8 x 10^{-5}, 14.0 x 10^{-5} M) Source: Mallinckrodt	$NaAsO_2$ was negative in the presence of Aroclor-induced rat S-9 mix; $NaAsO_2$ was positive for the three highest doses tested in the absence of S-9 mix; Na_2HAsO_4 was negative in the absence of an Aroclor-induced rat S-9 mix; Na_2HAsO_4 was positive at the three highest doses tested in the presence of an S-9 mix.	1. Chemical exposure was for 4 hours. 2. Only positive data were shown; negative results were only mentioned in the text. 3. The spontaneous mutation rate varied a great deal between experiments; between 2.5 and 17.9 mutants per 10^5 survivors. Most of the arsenic-induced mutant frequencies were between or just slightly above this spontaneous range. 4. The increases in mutant frequencies occurred at very low survival levels for both compounds; the highest doses caused 97% lethality. 5. At survivals above 10% none of the mutant frequencies exceeded the limits of the control mutant frequency range.

test protocols. Indeed, arsenic showed a dose-dependent suppression of mutations in strain WP44$_S$-NF. This strain carries a mutation (tif-1) which confers a thermally inducible SOS repair system (error-prone repair) resulting in normal spontaneous mutation at 30°C and a 20-fold increase in mutation at 42°C. The presence of sodium arsenite in the medium was sufficient to suppress this temperature-induced increase in the mutation frequency by one-half. This observation suggests that arsenic may inhibit SOS repair, a pathway which is highly important in bacterial mutagenesis. Such a repair pathway has not been detected in mammalian cells.

Rossman et al. (1980) offered explanations for the discrepancies between their results and those of Nishioka. First, they pointed out that the arsenic-induced mutation frequencies reported by Nishioka were all within the normal range of background. Second, they observed a 31-fold difference in cytotoxicity in the same bacterial strains. Third, they noted that Nishioka used an incorrect method for determining mutation frequency, basing his calculations on the initial size of the cell inoculum instead of on the final size of the plated population.

Arsenite and arsenate have also been found to be negative in the Ames Salmonella microsome assay (Lofroth and Ames, 1978). Although this work is reported only in abstract form, it may be assumed the assay was performed properly as Ames is a cited author.

Consideration of the above results alone, however, should not be taken as proof that inorganic arsenic is non-mutagenic. Bacterial systems are generally insensitive for the detection of metal mutagens because of the large amount of magnesium salts, citrate and phosphate in the minimal medium which prevent the entry of metal mutagens into bacterial cells (McCann et al., 1975).

6.1.2 Eukaryotic Microorganisms (Yeast, Fungi)

Sodium arsenite was tested for its activity in inducing reverse mutation and mitotic gene conversion in the yeast *Saccharomyces cerevisiae* (Singh, 1983). The diploid strain used in this study (D7) is auxotrophic for tryptophan, isoleucine, and valine, and measures gene conversion at the trp locus and reverse mutation at the ilv locus. Arsenite was inactive for gene conversion and weakly positive for mutation. This study was generally lacking in details and raw data were not shown.

6.1.3 Mammalian Cells In Vitro

Sodium arsenate was tested for the induction of forward mutations at the thymidine kinase locus in mouse L5178Y cells (Amacher and Paillet, 1980). Doses of arsenate were selected such that between 66 percent to 35 percent of cells survived a three-hour treatment. Arsenate was found not to increase the spontaneous mutation rate in these cells, while other known or suspected mutagenic metals (cadmium, nickel, and trans-platinum) were positive.

Using the same system, Oberly et al. (1982) tested sodium arsenate and sodium arsenite in the presence and in the absence of an Aroclor-induced rat S-9 mix. Sodium arsenite gave positive results at the three highest doses when tested in the absence of S-9 mix. In the presence of S-9 mix, the results were reported to be negative, although no data were presented. Sodium arsenate gave positive results at the three highest doses tested in the presence of S-9 mix. Again, without presenting data, the outcome in the absence of metabolic activation was reported to be negative. The authors concluded that these two arsenic compounds were weak mutagens. There are, however, several problems with this interpretation. First, the spontaneous mutation rate varied a great deal in these studies: between 2.5 and 17.9 mutants per 10^5 survivors. Most of the arsenic-induced mutant frequencies fell between or just slightly above this spontaneous range. Second, the increases in mutant frequencies occurred at very low survival levels for both compounds; the highest doses used caused 97 percent lethality. Thus, at survivals exceeding 10 percent none of the mutant frequencies exceeded the limits of the range of control mutant frequencies. Taking these observations into consideration, the authors' conclusions do not seem well supported.

Sodium arsenite was also tested for the induction of forward mutations at two loci in V79 cells (Chinese hamster lung fibroblasts) (Rossman et al., 1980). The two genes studied were hypoxanthine guanine phosphoribosyl transferase (HGPRT) and membrane-bound sodium potassium ATPase. Mutations in these two enzymes were enumerated through the observation of cell clones resistant to thioguanine or ouabain, respectively. Both the concentration of the chemical as well as the incubation times were varied; there was, however, no increase in the mutation frequency at either locus. The positive controls, UVC (ultraviolet light 254 mm) and methylnitronitrosoguanidine (MNNG) were effective in elevating the mutant frequency.

6.2 CHROMOSOMAL ABERRATION STUDIES

6.2.1 Chromosomal Aberrations In Vitro

A large number of studies indicate that both trivalent and pentavalent arsenic compounds are capable of breaking chromosomes in vitro (Table 6-2). One of the earliest reports on the clastogenic potential of arsenic was from a study performed with mitogenically stimulated human peripheral lymphocytes (Petres and Hundeiker, 1968). Although the test compound was reported to be Na_2HAsO_3, it is assumed the authors meant Na_2HAsO_4. Positive results were reported, although no actual raw data were presented.

Paton and Allison (1972) tested both sodium arsenite and sodium arsenate for the induction of chromosomal aberrations in human diploid fibroblasts and

TABLE 6-2. SUMMARY OF MUTAGENICITY OF ARSENIC: CHROMOSOME ABERRATIONS

Reference	Test System	Chemical Information	Results	Comments
Petres and Hundeiker, 1968 (paper in German, English summary)	Chromosome aberrations induced in human peripheral lymphocytes exposed in vitro.	Na_2HAsO_3 0.5 x 10^{-6} and 10^{6} g/ml of medium (assume Na_2HAsO_4 is meant) 1.6 x 10^{-6} M and 3.2 x 10^{-6} M	Positive at both doses.	1. No raw data were presented in this paper.
Paton and Allison, 1972	Chromosomal aberrations in human fibroblasts and human peripheral lymphocyte (HPL) cultures exposed in vitro.	$NaAsO_2$ (2.9 x 10^{-9} to 1.8 x 10^{-8} M) HPL. Na_2HAsO_4 (5.8 x 10^{-8} M) HPL. $NaAsO_2$ (2.9 x 10^{-9} to 5.8 x 10^{-8} M) fibroblasts. Source: British Drug House Chemical LTD. Solvent: H_2O	Positive for $NaAsO_2$ in HPL. Positive for $NaAsO_2$ in fibroblasts. Negative for Na_2HAsO_4 in HPL.	1. Incubation with chemical was for 48 hrs and 24 hrs for HPL and fibroblasts, respectively. 2. "Subtoxic" doses were used.
Larramendy et al., 1981	Induction of chromosomal aberrations in Syrian hamster embryo (SHE) cells and human peripheral lymphocytes (HPL) in vitro.	$NaAsO_2$ (7.7 x 10^{-6} M) in HPL and SHE. Na_2HAsO_4 (3.2 x 10^{-5} M) in HPL and SHE. Source: Chemical Services Inc. "100% pure" Solvent: Cell culture medium	Highly significant increase in the aberration frequency with both compounds in HPL and SHE.	1. Arsenite was about an order of magnitude more genotoxic than arsenate. 2. Both compounds induced mainly chromatid type aberrations. 3. Doses tested were generally non-cytotoxic. 4. Exposure to arsenic was for 24 and 48 hrs. in SHE and HPL, respectively.

TABLE 6-2. (continued)

Reference	Test System	Chemical Information	Results	Comments
Nakamuro and Sayato, 1981.	Chromosomal aberrations in cultured human peripheral lymphocytes (HPL) and fibroblasts.	As_2O_3 (3×10^{-7}, 12×10^{-7}, 36×10^{-7}M). $AsCl_3$ (6×10^{-7}, 24×10^{-7}, 72×10^{-7}M). $NaAsO_2$ (6×10^{-7}, 24×10^{-7}, 72×10^{-7}M). Na_2HAsO_4 (24×10^{-7}, 72×10^{-7}, 240×10^{-7}M). H_3AsO_4 (6×10^{-7}, 24×10^{-7}, 72×10^{-7}M). As_2O_5 (3×10^{-7}, 12×10^{-7}, 36×10^{-7}M). Source: Wako Pure Chemical Co. Solvent: H_2O	1. All arsenic compounds were clastogenic, all gave linear dose-response functions in lymphocytes. $NaAsO_2$ and H_3AsO_4 were positive with linear dose-response functions in fibroblasts. 2. Trivalent compounds were more potent than pentavalent compounds. The observed order for decreasing potency was: As_2O_3 > $AsCl_3$; Na AsO_2 >> Na_2HAsO_4 > H_3AsO_4; As_2O_5.	1. Treatment of lymphocytes was for the last 24 hours of culture. 2. Fibroblasts were exposed for 48 hrs. 3. All compounds induced more chromatid-type aberrations.

TABLE 6-2. (continued)

Reference	Test System	Chemical Information	Results	Comments
Nordenson et al., 1981	Chromosomal aberrations in peripheral lymphocytes of 3 normal volunteers - *in vitro* exposure.	NaAsO$_2$ (77 x 10^{-8}, 150 x 10^{-8}, 230 x 10^{-8}, 310 x 10^{-8} M). Na$_2$HAsO$_4$ (77 x 10^{-8}, 150 x 10^{-8}, 230 x 10^{-8}, 310 x 10^{-8} M). Source: Merck Solvent: Normal saline	NaAsO$_2$ caused a dose-related increase in gaps and chromatid breaks. Na$_2$HAsO$_4$ was negative.	1. Arsenic added 24 hours after initiation of culture, harvested at 72 hours. 2. Only scored gaps, chromatid and chromosome breaks. 3. Also examined G$_0$ effects: 24 hr. exposure without PHA, remove arsenic, add PHA, harvest after 48 hours; examined "NaAsO$_4$" - do not know what compound was really tested. 4. Reason for negative result with arsenate is that a sufficiently high dose was not tested. Should have increased concentration by an order of magnitude.

TABLE 6-2. (continued)

Reference	Test System	Chemical Information	Results	Comments
Wan et al., 1982	Induction of chromosome aberrations *in vitro* in CHO cells and human peripheral lymphocytes (HPL).	$NaAsO_2$ (10^{-6}, 5×10^{-6}, 10^{-5} M) CHO; (0.5×10^{-6}, 10^{-6}, 5×10^{-6} M) HPL Na_2HAsO_4 (5×10^{-5}, 8×10^{-5}, 10^{-4} M) CHO. Purity: Analytical grade Source: Fisher Scientific Co. Solvent: H_2O	Dose-related increase for both compounds in CHO. $NaAsO_2$ gave dose-related increase in HPL.	1. Trivalent was 5 to 10 x more potent than pentavalent. 2. All doses tested were inhibitory to CHO cell growth. 3. Arsenic present for entire culture period (22 hrs.) in CHO cells, and final 48 hrs of culture in HPL.
Poma et al., 1981b	Induction of chromosomal aberrations in mouse bone marrow and spermatogonia *in vivo*.	As_2O_3, single I.P. injection (4, 8 and 12 mg As/kg). Solvent: H_2O	Negative in bone marrow. Negative in spermatogonia.	1. Mouse strain: Swiss Albino 2. Sex: male. 3. Age: 12 weeks 4. Number of animals per dose group: 4-8. 5. Cells per animal: 100. 6. Sacrifice times: 12, 24, 36, 48 hrs post injection.

mitogenically stimulated human peripheral lymphocytes. Sodium arsenite gave positive results in both cell types while sodium arsenate was tested only in lymphocytes and was reported to be negative. Since arsenate (which most investigators find to be considerably less potent than arsenite) was tested at a lower concentration, the negative result is not surprising.

In a more recent study, the effects of sodium arsenite and sodium arsenate on chromosomal aberration frequencies were examined in cultured human peripheral lymphocytes and in Syrian hamster embryo cells (Larramendy et al., 1981). Both compounds gave highly significant increases in chromosomal aberrations in both cell types. Arsenite was more potent by approximately an order of magnitude, and both compounds induced mainly chromatid-type aberrations.

Nakamuro and Sayato (1981) tested six arsenic compounds for the induction of chromosomal aberrations in cultured human peripheral lymphocytes and in human skin fibroblasts. All arsenic compounds tested were clastogenic and all gave linear dose-response functions in lymphocytes. Trivalent compounds were more potent than pentavalent compounds. The observed order for decreasing potency was $As_2O_3 > AsCl_3$; $NaAsO_2 >> Na_2HAsO_4 > H_3AsO_4$; As_2O_5 (Figure 6-1). $NaAsO_2$ and H_3AsO_4 were positive, showing linear dose-response functions in fibroblasts. All compounds induced predominantly chromatid-type aberrations.

In another study, the effects of sodium arsenite and sodium arsenate on the induction of chromosomal aberrations in human peripheral lymphocytes exposed *in vitro* were examined using two protocols (Nordenson et al., 1981). In the first protocol, cells were mitogenically stimulated with PHA, arsenic was added after 24 hours and cells were harvested after 72 hours (48-hour arsenic exposure). Using this method, sodium arsenite was found to increase the frequency of chromatid breaks and gaps in a dose-dependent fashion. Sodium arsenate was reported to be negative, but these investigators employed

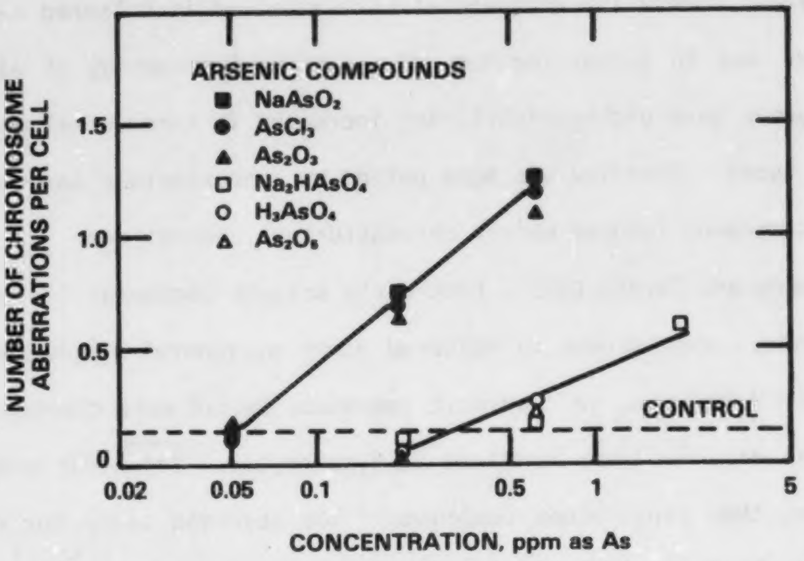


Figure 6-1. The induction of chromosome aberrations in cultured human peripneral lymphocytes by six arsenic compounds.

Source: Nakamuro and Sayato (1981).

the same concentrations of arsenate as were tested for arsenite. It is possible that higher doses of arsenate would also have yielded positive results. In the second protocol, G_0 (unstimulated) lymphocytes were exposed to arsenic for 24 hours, and the cells were washed, stimulated with PHA and harvested after 48 additional hours. Using this protocol the authors reported that "$NaAsO_4$" was negative. One may assume that the authors meant $NaAsO_2$, since having observed a lack of activity for arsenate in the first protocol, it would seem unwarranted to test it in the second. If the test compound was indeed arsenite, these observations suggest that arsenic has its effect through some influence on DNA synthesis and not by directly damaging DNA.

In the most recent study on in vitro clastogenic effects of arsenic compounds, sodium arsenite and sodium arsenate were tested in CHO cells and human peripheral lymphocytes (Wan et al., 1982). Both compounds showed dose-dependent increases in aberrations in CHO cells; arsenite was also positive in lymphocytes. Again, trivalent arsenic was 5 to 10 times more potent than pentavalent arsenic.

6.2.2 Chromosomal Aberrations In Vivo

There is only one report in the literature (excluding abstracts) in which the clastogenic effects of arsenic were tested in a mammalian species in vivo (Poma et al., 1981b). These authors examined chromosomal aberration frequencies in mouse bone marrow cells and spermatogonia after administration of As_2O_3 (4, 8 and 12 mg of arsenic/kg). This trivalent form of arsenic was previously shown to be the most potent form in vitro (Nakamuro and Sayato, 1981). Four to 8 animals were included in each dose group; animals were killed at 12, 24, 36 and 48 hours after exposure and 100 metaphase cells per animal were scored. This represents an acceptable protocol, and the study appears to have been properly performed. There was no significant increase in aberrations in either bone marrow or spermatogonia following arsenic treatment. An important question in this study is whether sufficiently high doses were tested. The LD_{50} for arsenic trioxide in rats is 104 mg As/kg; thus, the highest dose used in this mouse study is only 13 percent of the rat LD_{50}. One can crudely compare the doses of arsenic trioxide which were effective _in vitro_ to those used in this study by simply equating the test animal to an equivalent volume of water. Such a comparison indicates the mice were "1.6×10^{-4} M" effective for arsenic while the same compound was effective for inducing aberrations _in vitro_ at approximately 0.7 to 7.0×10^{-4} M arsenic. Clearly this represents a very crude comparison since intact animals can bind, compartmentalize and excrete a toxicant while cells in culture cannot. Thus, it is not clear whether higher doses should have been tested in this study.

6.2.3 Chromosomal Aberrations and SCE in Cells of Arsenic-Exposed Human Beings

The majority of studies indicate that people exposed to arsenic due to occupation or pharmaceutical treatment display increased levels of chromosomal aberrations and SCE in peripheral lymphocytes (Table 6-3). It should be noted that most of the studies which have examined aberrations and SCE are poor in quality due either to errors in experimental protocol, misclassification of aberrations, poor choice in controls and/or not performing studies in a blind fashion.

Beckman et al. (1977) examined the frequency of chromosomal aberrations in peripheral lymphocytes of nine smelter workers who were exposed to arsenic. The levels and the forms of arsenic to which the workers were exposed were not indicated. A statistically significant increase in aberrations was found in the exposed population. There are several problems with this study. First, the authors use an incorrect definition of gaps. They state: "Gaps are commonly defined as achromatic regions exceeding the width of a chromosome." This, in

TABLE 6-3. SUMMARY OF MUTAGENICITY OF ARSENIC: CHROMOSOMAL ABERRATIONS AND SCE IN CELLS OF EXPOSED HUMANS

Reference	Test System	Chemical Information	Results	Comments
Beckman et al., 1977	Chromosomal aberrations in lymphocytes of 9 smelter workers exposed to arsenic.	Arsenic from smelter	Positive, six-fold increase in percent aberrant cells; eight-fold increase in aberrations per cell.	1. Authors used an incorrect definition of gaps. 2. Cells cultured too long (72 hours). 3. Cells scored per individual were unknown. 4. Number of control individuals and cells per control individual were not specified. 5. Some workers were also exposed to lead and selenium.
Nordenson et al., 1978	Chromosomal aberrations in lymphocytes of 33 smelter workers.	Arsenic from smelter	Positive	1. Divided workers into high exposure (18 individuals), medium exposure (11), low exposure (4) and new employees (6). 2. The controls were an unspecified number of apparently historical controls. 3. No dose effect. 4. Wrong definition of gaps. 5. Cell culture time too long. 6. Large difference between controls and new employees.

TABLE 6-3. (continued)

Reference	Test System	Chemical Information	Results	Comments
Burgdorf et al., 1977	Compared SCE and chromosomal aberrations in lymphocytes of 6 patients treated with arsenic, with results from 44 control individuals.	All treated with Fowler's solution (1% $KAsO_2$ in H_2O), for time periods between 4 and 27 years. Five of these individuals had not been treated for at least 24 years.	No difference in frequencies of chromosomal aberrations. SCE frequencies were increased more than 2-fold in arsenic-treated patients.	1. Cultured cells for 72 hours - too long for examination of aberrations. 2. No indication that study was scored blind. 3. Patients exposed to other agents (e.g. 4000 rad x-irradiation, 5000 rad x-irradiation, fluoroscopy twice weekly while receiving arsenic, 4 had asthma).
Petres et al., 1977	Chromosome aberrations in lymphocytes of 31 individuals exposed to arsenic.	Arsenic exposure related to treatment for psoriasis or associated with vine-growing.	Positive, nine-fold increase in aberrations among arsenic-exposed individuals.	1. 31 exposed individuals, 31 controls. 2. Cell culture time too long (72 hours). 3. Scored both gaps and achromatic lesions - same aberration. 4. Scored secondary constrictions - not generally considered an aberration. 5. No indication of the number of cells per person scored. 6. Saw a 5-fold increase in aneuploidy among arsenic-exposed individuals, always hypodiploidy, which suggests an artifact.

TABLE 6-3. (continued)

Reference	Test System	Chemical Information	Results	Comments
Nordenson et al., 1979	Chromosome aberrations and SCE in peripheral lymphocytes of 8 psoriatic patients treated with arsenic.	Total arsenic dose per individual varied from a low of 300 mg up to 1200 mg. Most had not been treated for 15 years.	Psoriatic patients treated with arsenic showed significantly higher (three-fold) levels of aberrations compared to control psoriatics. Psoriatic patients treated with arsenic had the same levels of SCE as controls.	1. Slides were scored blind. 2. Investigators controlled for age and sex but not for ethanol use, smoking, drug use, or infection. 3. The increases in aberrations were not dose-related. 4. Cultured cells for 72 hours: too long for aberrations. 5. Scored too few cells for SCE in control patients; provided total number of cells scored, but number of cells per individual was unknown.

TABLE 6-3. (continued)

Reference	Test System	Chemical Information	Results	Comments
Wen et al., 1981	SCE in peripheral lymphocytes of 13 Blackfoot disease patients (arsenic-exposed) and 13 controls.	Exposure to arsenic from well water; years of use ranged from 15 to 58. Most had not consumed contaminated water for 12 years.	Arsenic-exposed individuals showed small but statistically higher SCE level. Ranges for arsenic-exposed (9.1 to 17.5 SCE/cell) and controls (7.8 to 12.0 SCE/cell) showed considerable overlap.	1. Patients exposed to a large number of other drugs, including pantopan, scopolamine, promethazine hydrochloride, meperidine, ketamine hydrochloride, Pyrabital, Valium, ampicillin, Kanamycin, Sedes, diphenhydramine hydrochloride, Kolantyl, Complamin, Venostatin, Ulcerin, metoclopramide, Sulpyrin, chloramphenicol, reserpine, Bartropine, dextromethorphan, furosemide, and Hustazol. 2. All patients had undergone leg or foot amputation.

fact, is a definition for a break. A gap is commonly defined as an achromatic lesion not exceeding the width of a chromatid arm. Second, the total number of cells examined are given but the cells per individual worker, cells per individual control, and total number of control individuals are not listed. Third, these investigators cultured the lymphocytes for 72 hours, a duration 24 hours longer than optimum, since most chromosome aberrations are cytotoxic, and cells with such lesions generally do not survive more than a single cell cycle. After 72 hours of culture, the majority of cells have divided three times, eliminating many damaged cells. Clearly, however, this type of error tends to underestimate the frequency of aberrations. Finally, workers were also exposed to other potentially clastogenic agents such as lead and selenium.

A report one year later by the same group (Nordenson et al., 1978) extended this study to a total of 33 smelter workers. In this study individuals were grouped into high exposure (18 people), medium exposure (11), low exposure (4) and new employees (6). The groupings were based on the nature of the job; quantitative measurements of exposure were not performed. The workers were reported to have a higher level of chromosome aberrations than did the controls. However, the controls were of an unspecified number and, apparently, historical, the definition of gaps was incorrect, cells were cultured for 72 hours, and large differences were found between historical controls and new employees. Due to these problems, the positive findings reported are questionable.

Burgdorf et al. (1977) compared SCE and chromosomal aberration frequencies in lymphocytes of six patients treated with arsenic (for asthma, psoriasis or anxiety) to results from 44 control individuals. All patients had been treated with Fowler's solution (1% $KAsO_2$ in water) for time periods varying between 4 and 27 years. Five of these individuals had not been treated for at least 24 years. The authors found no difference in frequencies of chromosomal aberrations, but reported a more than 2-fold higher SCE frequency in arsenic-treated patients. Each of the patients had an SCE level which was significantly higher than the mean level for controls. Problems with this study included a 72-hour culture period for aberrations, no indication that slides were scored "blind," and the fact that patients were exposed to other agents (4000 rad x-irradiation, 5000 rad x-irradiation, fluoroscopy twice weekly concomitant to arsenic exposure).

In a similar study, Petres et al. (1977) compared chromosome aberrations in lymphocytes of 31 arsenic-exposed people to 31 controls. Arsenic exposure resulted from treatment for psoriasis or was associated with vine-growing. The levels and forms of arsenic exposure were not indicated. A statistically significant increase in aberrations was found in the exposed group. These authors also reported a five-fold increase in aneuploidy among arsenic-exposed individuals. Problems in this study include: (1) a 72-hour cell culture period; (2) no indication of the number of cells per person scored; (3) scoring both gaps and achromatic lesions, identical endpoints; (4) scoring secondary constrictions, which are generally not considered to be aberrations; and (5) all aneuploid cells were hypodiploid, which suggests an artifact.

Nordenson et al. (1979) compared chromosome aberrations and SCE in peripheral lymphocytes of 8 arsenic-treated psoriatic patients to 8 untreated psoriatic controls. Total arsenic dose per individual varied from a low of 300 mg up to 1200 mg. Most had not been treated for 15 years. Psoriatic patients treated with arsenic showed significantly higher levels of aberrations compared to controls, but showed similar frequencies of SCE. The increase in aberrations was not related to the total arsenic dose. Although these authors controlled for age and sex, there was no apparent control for smoking, drug use or ethanol consumption. An average of 15 cells per individual were scored for SCE; generally, 20 cells per individual is considered the acceptable minimum. As in the other studies, the cells were cultured for 72 hours.

Frequencies of SCE were compared in 13 patients exposed to arsenic in drinking water to SCE frequencies in 13 control individuals (Wen et al., 1981). All patients displayed symptoms of Blackfoot disease, a peripheral vascular disease resulting from chronic arsenic intoxication. Patients had used contaminated water for 15 to 58 years; most had switched to uncontaminated tap water 12 years before this study. Arsenic-exposed individuals showed a small but statistically higher SCE level. Ranges for arsenic-exposed (9.1 to 17.5 SCE/cell) and controls (7.8 to 12.0 SCE/cell) showed considerable overlap. Sources of error in this study are large since the arsenic-exposed group had also been exposed to over 20 different pharmaceuticals.

6.3 OTHER STUDIES INDICATIVE OF MUTAGENIC DAMAGE

6.3.1 Preferential Killing of Repair-Deficient Bacterial Strains

Arsenic compounds have generally been found to preferentially kill bacteria that carry particular mutations in DNA repair pathways (Table 6-4). Both sodium arsenite and sodium arsenate gave positive results in the *B. subtilus* "rec assay" (Nishioka, 1975). In this assay, test chemicals are spotted onto filter discs and the discs are placed at the centers of a bacterial lawn composed of rec^+ cells (H17) or rec^- cells (M45, deficient in recombinational DNA repair). The size of the zone of growth inhibition is measured for both strains, with a differential response (more killing of M45) indicative of a DNA-damaging agent. Sodium arsenite was reported to give a moderately positive response while sodium arsenate was reported to give a weakly positive response.

The results for sodium arsenate were reproduced in the *B. subtilus* rec assay and positive responses for four other arsenic compounds (As_2O_5, As_2O_3, $AsCl_3$, $3AsO_5$) have been reported (Kanematsu et al., 1980). These authors indicated that all arsenic compounds gave "strongly positive" responses (greater than 6 mm growth difference).

TABLE 6-4. MUTAGENICITY OF ARSENIC: ASSAYS INDICATIVE OF DNA DAMAGE OR REPAIR

Reference	Test System	Chemical Information	Results	Comments
Nishioka, 1975	Rec assay in B. subtilus. Strains: H_{17} (rec$^+$, arg$^-$, trp$^-$) M45 (rec$^-$, arg$^-$, trp$^-$	$NaAsO_2$ (0.05 M) Na_2HAsO_4 (0.05 M) 0.05 ml aliquot spotted onto filter disc. Source: Nakarai Chemical LTD. Solvent: H_2O	$NaAsO_2$ moderate positive response in rec assay. Na_2HAsO_4 weak positive response in rec assay.	
Kanematsu et al., 1980	Implication of DNA damage through the measurement of growth of normal (rec+) and recombination-repair-deficient (rec$^-$) B. subtilis, "rec assay." Strains: H_{17} (rec$^+$, agr$^-$, trp$^-$) M45 (rec$^-$, arg$^-$, try$^-$).	As_2O_5 (0.05 M) As_2O_3 (0.05 M) $AsCl_3$(0.03 ml) $3AsO_5$ (0.05 M) Na_2HAsO_4 (0.1 M) 0.05-ml portions of aqueous solutions at the indicated concentration are applied to the paper disc. Source: Marvichi Chemical LTD. Solvent: H_2O	All arsenic compounds gave strong positive responses. (Greater than 6-mm difference in growth).	1. Plates kept at 4°C for 24 hrs. prior to incubation at 37°C. This "cold incubation" increases the sensitivity of the assay 20 to 50 times.

TABLE 6-4. (continued)

Reference	Test System	Chemical Information		Results	Comments
McCarroll et al., 1981	E. coli micro-suspension assay. Measures preferential killing of repair-deficient strains. Strains WP2 (wild type) WP2 UVRA (uvrA$^-$) CM611 (lexA$^-$, uvrA$^-$) WP67 (polA$^-$, (uvrA$^-$) WP100 (recA$^-$, uvrA$^-$) WP3110 (thy$^-$) P3478 (polA$^-$, thy$^-$)	Na_2HAsO_4 $NaAsO_2$	500, 250, 125, 62.5, 31.3, 15.6, 7.8, 3.9, 1.9, 0.98, 0.5, 0.2 (μg/well). Same	Na_2HAsO_4 was positive at 130 μg/well in strain WP100. $NaAsO_2$ was positive at 63 μg/well in strain CM 611. Results in all other strains were negative.	1. The total volume per well is not clearly stated, thus the molarity of the arsenic solutions can not be calculated 2. Strain WP100 is deficient in excision repair and in postreplicational repair (both SOS error-prone and recombinational). 3. Strain CM611 is deficient in excision repair and in post-replicational error-prone (SOS) repair.
Nordenson et al., 1981	SCE in peripheral lymphocyes, *in vitro* exposure.	Source: Merck Solvent: Normal saline $NaAsO_2$ (390 x 10^{-8} M)		Positive SCE response (1.5-fold increase).	1. Added arsenic to cultures 24 hours after initiation of culture, harvested at 72 hours.

TABLE 6-4. (continued)

Reference	Test System	Chemical Information	Results	Comments
Larramendy et al., 1981	Induction of SCE in Syrian hamster embryo (SHE) cells and human peripheral lymphocytes (HPL) in vitro.	$NaAsO_2$ (7.7 x 10^{-7}, 1.9 x 10^{-6} M) in HPL. Na_2HAsO_4 (1.6 x 10^{-5}, 3.2 x 10^{-5} M) in HPL. $NaAsO_2$ (3.8 x 10^{-7}, 3.8 x 10^{-6}, 7.7 x 10^{-6} M) SHE. Na_2HAsO_4 (3.2 x 10^{-6}, 8 x 10^{-6}, 1.6 x 10^{-5}, 3.2 x 10^{-5} M) SHE. Source: Chemical Services Inc. "100% pure" Solvent: Cell culture medium	Dose-related increases in SCE with both compounds in both HPL and SHE (two-fold increases).	1. Arsenite was about an order of magnitude more genotoxic than arsenate. 2. Doses tested were generally non-cytotoxic. 3. Both compounds were less efficient in inducing SCE in human whole blood cultures than in purified lymphocyte cultures. 4. Exposure to arsenic was for 24 and 48 hrs. in SHE and HPL, respectively.
Andersen, 1983	Sister chromatid exchange in cultured human lymphocytes (3 donors); and in P388D cells (continuous line of macrophage cells derived from DBA/2 mouse).	$NaAsO_2$ (10^{-6}, 10^{-7}, 10^{-8} M) P388D cells. Na_2HAsO_4 (10^{-5}, 10^{-6}, 10^{-7} M) P388D cells. $NaAsO_2$ (10^{-6} M) human lymphocytes. Solvent: H_2O	Both compounds negative in P388D cells. $NaAsO_2$ positive in lymphocytes from all three donors.	1. Chemical exposure was 48 hrs. in P388D cells. 2. Chemical exposure 24, 48, and 72 hrs. in lymphocytes. 3. Results in lymphocytes variable between donors, ranging from 30% to 117% increases in SCE frequencies for the three individuals.

TABLE 6-4. (continued)

Reference	Test System	Chemical Information	Results	Comments
Crossen, 1983	Sister chromatid exchange in cultured peripheral lymphocytes from 4 normal human volunteers.	Na_2HAsO_4 (1.34×10^{-5}, 2.68×10^{-5} M) $NaAsO_2$ (7.69×10^{-7}, 1.54×10^{-6} M) Source: BDH Chemicals Solvent: H_2O	Na_2HAsO_4 - 1 donor increased SCE, 2 donors unchanged SCE, 1 donor decreased SCE. $NaAsO_2$ - 2 donors increased SCE, 2 donors unchanged. Both compounds negative when cells treated during G_o and not present during replication.	1. Results suggest that arsenic does not damage DNA but may interfere with DNA replication. 2. Even positive responses were generally not dose-related. 3. Large variation in baseline SCE levels between individuals. 4. Both compounds present during final 48 hrs of culture.

TABLE 6-4. (continued)

Reference	Test System	Chemical Information	Results	Comments
Ohno et al., 1982	SCE induction in DON Chinese hamster cells in vitro.	$NaAsO_2$ (7.7 x $10.^{-6}$ M) As_2O_5 (1.4 x 10^{-5} M) Na_2HAsO_4 (5.4 x 10^{-6} M)	All compounds gave a significant increase in the SCE frequency (1.5-fold, 1.6-fold and 1.5-fold, respectively).	1. Chemical exposure was for 28 hrs. 2. Doses tested caused a 50% inhibition in cell growth.
Wan et al., 1982	Induction of SCE in CHO cells in vitro.	$NaAsO_2$ (10^{-6}, 10^{-5} M) Purity: analytical grade. Source: Fisher Scientific Co. Solvent: H_2O	Dose-related increase in SCE (2.4-fold increase).	1. Arsenic present for entire culture period (24 hrs).

TABLE 6-4. (continued)

Reference	Test System	Chemical Information	Results	Comments
Tkeshelashvili et al., 1980	Fidelity of DNA synthesis components: 1. *E. coli* DNA polymerase. 2. Artificial DNA template poly-[d(A-T)]. 3. Radiolabeled nucleotide triphosphates	Na_2HAsO_4 (0.1, 0.5, 1.0., 5.0 mM) As_2O_5 (0.1, 0.5, 2.5, 10.0 mM) Source: Either ICN Corp. or J.T. Baker Purity: 99.9%	Neither arsenic compound affected the "natural" error rate.	1. Other metals such as chromium increase the error rate in this system. 2. This is a very artificial system, i.e., the polymerase is a bacterial-repair enzyme, the DNA is an artificial template containing only two nucleotide bases and the system is carried out entirely *in vitro*.
Fornace and Little, 1979	Induction of DNA-protein crosslinking and single-strand DNA breaks in mouse C3H/10T 1/2 cells (alkaline elution).	$NaAsO_2$ (1 mM) Solvent: Cell culture medium	No crosslinks detected. Positive for single-strand breaks (equivalent to 120 rad of X-ray)	1. Cells were treated for 1 hour; only 2.2% of cells survived this treatment.

Sodium arsenate and sodium arsenite were also found positive in the *E. coli* microsuspension test (McCarroll et al., 1981). In this assay, a variety of *E. coli* strains carrying mutations for DNA repair are incubated in multiwell microplates with a serially-diluted test compound. The highest dose at which growth is evident is compared in wild type and DNA repair-deficient strains. Again, a differential sensitivity to growth inhibition is indicative of DNA damage. Sodium arsenate was found to preferentially inhibit the growth of strain WP100 (recA^{-}, uvrA^{-}) compared to wild type cells. This strain is deficient in excision as well as recombinational repair. Sodium arsenite was positive in strains WP100 and CM611 (lexA^{-}, uvrA^{-}). This latter strain is deficient in excision repair and is not inducible for post-replicational error-prone (SOS) repair. Negative results were seen in strains which are only deficient in excision repair, implying that arsenic-induced damage is not repaired by this pathway or that arsenic has no effect on enzymes which repair spontaneous damage through excision repair.

6.3.2 DNA Damage in Mammalian Cells In Vitro

Fornace and Little (1979) examined the effects of sodium arsenite on the induction of DNA-protein crosslinks and single-strand DNA breaks in mouse C3H/10T 1/2 cells using alkaline elution (Table 6-4). This technique measures the rate at which radiolabeled DNA passes through a polyvinyl chloride filter having a defined pore size. Treatment of cells for one hour did not induce DNA-protein crosslinks but did cause a significant increase in single-strand breaks. The level of breakage observed was equivalent to treating the cells with 120 rad of x-ray. It should be noted that the treatment protocol was highly toxic with only 2.2 percent of cells surviving the treatment.

6.3.3 Fidelity of DNA Synthesis

Some metals, such as chromium, are thought to be mutagenic by interfering with a DNA polymerase-editing function. As a result they increase the rate at which "errors" are made in DNA replication. Loeb and co-workers have designed an *in vitro* system that measures the rate at which particular nucleotides are inappropriately incorporated into DNA (Sirover and Loeb, 1976; Tkeshelashvili et al., 1980) (Table 6-4). In this system, the DNA template is a repeating sequence of adenine and thymidine (poly[d(A - T]) which is incubated with *E. coli* DNA polymerase I, and ^{32}P-labelled dTTP and ^{3}H-dGTP. By monitoring the ratio of the two isotopes in acid precipitable material one can quantify the frequency with which an error in incorporation is made. Neither sodium arsenate nor arsenic pentoxide (As_2O_5) affected the "natural" error rate. Chromium was shown to be positive in this assay. It should be cautioned that this is a very artificial system, i.e., the polymerase is a bacterial-repair enzyme, the DNA is an artificial template containing only two nucleotide bases, and the system is carried out entirely *in vitro*.

6.3.4. Induction of Sister Chromatid Exchanges (SCE) In Vitro

Arsenic compounds have been extensively tested for the induction of SCE in a variety of cell types *in vitro*. Almost without exception the results are positive (Table 6-4). Significant increases in SCE frequencies were shown in human peripheral lymphocytes treated with sodium arsenite (Nordenson et al, 1981; Larramendy et al., 1981; Anderson, 1983; Crossen, 1983) and sodium arsenate (Larramendy et al., 1981; Crossen, 1983). The studies by Crossen, however, suggested that individuals may vary significantly in their susceptibility to arsenic-induced SCE. Crossen noted that arsenite increased the SCE frequency in cells from two donors but had no effect in cells from two other donors. Arsenate increased the SCE frequency in one donor, had no effect in two other donors and slightly decreased the SCE frequency in a fourth donor. Crossen also used a protocol in which cells were exposed to arsenic in G_0 (unstimulated), washed and then cultured for SCE analysis. The results were similar to those seen for chromosomal aberrations (Nordenson et al., 1981), i.e., neither arsenite nor arsenate increased SCE levels when treatment occurred during a nonreplicative phase. Again, these results indicate that arsenic must be present during DNA synthesis (SCE occur during S phase) to have an effect.

Arsenic has also been tested for SCE induction in Chinese hamster lines *in vitro*. Wan et al. (1982) reported a dose-related increase in SCE in CHO cells exposed to sodium arsenite, while Ohno et al. (1982) reported significant increases in the SCE frequency in DON cells exposed to single doses of sodium arsenite, sodium arsenate, and arsenic pentoxide.

6.4 INTERACTIONS OF ARSENIC WITH OTHER MUTAGENS

6.4.1. Bacterial Studies

The effects of sodium arsenite (1 mM) on the frequencies of UVC-induced mutations were measured in *E. coli* strain WWP_2 (trp^-, $uvrA^-$), a strain deficient in excision repair (Rossman et al., 1977). Arsenite was found to reduce the number of UVC-induced mutations by 30 to 40 percent. This observation suggests that arsenic may interfere with SOS (error prone) DNA repair, and may, therefore, be a bacterial antimutagen (Table 6-5).

In a later study, Rossman (1981) further examined the effect of sodium arsenate and sodium arsenite on frequencies of UVC-induced mutations. In *E. coli* WP2, which is proficient in all repair pathways, arsenite increased UVC mutagenesis at doses up to 0.25 mM and then decreased the mutation frequency at higher doses. The increase in mutation was seen only in excision repair-proficient strains. These observations suggest that arsenic interferes with excision repair at low doses and interferes with SOS repair at higher doses.

Fong et al. (1980) examined the effect of sodium arsenate on the frequency of UVC-induced single-strand DNA breaks in *E. coli* (Table 6-5). The presence

TABLE 6-5. SUMMARY OF MUTAGENICITY OF ARSENIC: INTERACTIONS WITH MUTAGENS

Reference	Test System	Chemical Information	Results	Comments
Rossman et al., 1977	Interaction of UVC and arsenic in *E*. *coli* for mutation. Strain WWP_2 (trp^-, $uvrA^-$).	$NaAsO_2$ (1 mM) plus 1.25, 5.0, and 10 J/m^2 UVC	$NaAsO_2$ reduced the number of UVC-induced mutants by 30 to 40 %.	1. Suggests that arsenic interferes with SOS repair. 2. May not be relevant to mammals since this type of repair has not been found.
Rossman, 1981	Interaction of arsenic and UVC in the induction of mutations of *E*. *coli*. Strains: WP2 (trp^-) $WP2_S$ (trp^-, $uvrA^-$) WP6 (trp^-, $polA1^-$) WP5 (trp^-, $lexA^-$) WP10 (trp^-, $recA^-$)	$NaAsO_2$ (0.1, 0.25, 0.5, 0.75 mM) plus 5.6 J/m^2 in WP2. Na_2HAsO_4 (0.1, 0.3, 0.5 mM) plus 5.6 J/m^2 in WP2. $NaAsO_2$ (0.2 mM) plus 5.6 J/m^2 in WP2 0.6 J/m^2 in $WP2_s$ 1.2 J/m^2 in WP6 0.6 J/m^2 in WP5 0.6 J/m^2 in WP10 Source: Fisher Chemical Co.	Arsenite increased UVC mutagenesis in WP2 at doses up to 0.25 mM and then decreased mutation frequency at higher doses. This increase was not seen with arsenate. The effect with arsenite was seen only in excision repair-proficient strains.	1. Observations indicate that arsenic may interfere with excision repair at lower doses and with SOS repair at higher doses.

TABLE 6-5. (continued)

Reference	Test System	Chemical Information	Results	Comments
Fong et al., 1980	Induction of single-strand (S.S) DNA breaks in *E. coli* after exposure to UVC light (alkaline sucrose sedimentation). Strains used: WP2-wild type WP6-(polA), lacks DNA polymerase used in repair WP2-(uvrA), deficient in excision repair.	$NaAsO_2$ (1,2,3,4, 5 mM)	1 mM arsenic inhibited the induction of S.S. breaks in WP2 and WP6 after exposure to UVC (40 J/m^2 and 10 J/m^2, respectively). 3 mM arsenic prevented the rejoining of S.S. breaks after UVC (5 J/m^2) in WP2 (uvrA) cells.	1. Arsenic prevents enzymatic incision in DNA after exposure to ultraviolet light. 2. Authors speculate this may result from arsenic-induced depletion of cellular ATP. 3. Arsenic may inhibit post-replicational repair since it prevented rejoining of S.S. breaks in cells deficient in excision repair.
Jung et al., 1969	Inhibition of unscheduled DNA synthesis (UDS) in human epidermal cells seen in biopsies derived from seven normal volunteers after UV irradiation.	Na_2HAsO_4 (10^{-5}, 10^{-6} M)	Arsenic inhibited UVC-induced UDS.	1. Tissue biopsies were incubated with arsenic for either 1 hr or 4 hrs prior to UVC exposure. 2. There were no dose- or incubation time-related effects on inhibition of UVC-induced UDS. 3. Actual numbers were not shown.

TABLE 6-5. (continued)

Reference	Test System	Chemical Information	Results	Comments
Rossman, 1983 (progress report)	Mutation in V79 cells. Interaction of UVC and arsenite.	Arsenite	No interaction	1. No data provided.
Poma et al., 1981a (abstract)	Interaction between arsenic and ethyl methanesulfonate in the induction of chromosome aberrations *in vivo* in Swiss-albino mice: bone marrow, and spermatogonia.	Arsenic (12 mg As/kg) EMS (200 mg/kg) pre- or post-treatment.	No interaction	1. No experimental details, work described in an abstract.
Sram, 1976	Chromosomal aberrations in bone marrow cells of ICR mice. Each dose group consisted of 5 female mice. Animals sacrificed 24 hours after a single dose of TEPA, scored 250 metaphases/group (assume 50/animal).	$NaAsO_2$ in drinking water for 8 weeks. 10 mg/l (7.7×10^{-5} M) 100 mg/l (7.7×10^{-4} M) TEPA 2 mg/kg	$NaAsO_2$ alone did not appear to induce aberrations although the spontaneous rate was not clearly indicated. The lower dose of arsenic in drinking water did not affect the frequency of TEPA-induced aberrations. The higher dose of arsenic resulted in a synergistic increase in the aberration rate.	1. This study was lacking in experimental details.

TABLE 6-5. (continued)

Reference	Test System	Chemical Information	Results	Comments
Sram, 1976	Interaction of $NaAsO_2$ and TEPA for dominant lethals in ICR mice; each treated and control group consisted of 20 males and 40 females/week. Mating continued for 3 to 8 weeks after the application of TEPA.	$NaAsO_2$ in drinking water for 8 weeks/generation, males were exposed for 4 generations prior to mating. 10 mg/l (7.7 x 10^{-5} M) 100 mg/l (7.7 x 10^{-4} M) TEPA 2 mg/kg	$NaAsO_2$ alone did not appear to induce dominant lethals, although it is not clear what the spontaneous rate was. There appeared to be a positive interaction between TEPA and the higher exposure to arsenic.	1. This study was badly lacking in experimental details. 2. It was not indicated at what time after TEPA administration the increase in dominant lethals was seen, thus it is not known which germ cell stage was affected.

of strand breaks in DNA was measured by alkaline sucrose sedimentation. One mM arsenite was found to inhibit the induction of strand breaks in WP2 (wild type) and WP6 polA (lacks DNA polymerase I) after exposure to UVC. The presence of 3 mM arsenate in growth medium prevented the rejoining of strand breaks after UVC exposure in WP2 uvrA cells (excision repair-deficient). The authors interpreted these observations to suggest: (1) that arsenic prevents enzymatic incision of DNA after UVC exposure (UVC alone does not cause strand breaks), and (2) that arsenic may inhibit post-replicational repair since it prevented rejoining of breaks in cells deficient in excision repair. The authors further speculated that the inhibition of enzymatic incision may result from arsenic-induced cellular depletion of ATP.

6.4.2 Mammalian Cells In Vitro

The interaction of sodium arsenate and UVC on levels of unscheduled DNA synthesis (UDS) was measured in cells from human skin biopsies taken from seven volunteers (Jung et al., 1969; Table 6-5). Arsenate was tested at two concentrations (10^{-5}, 10^{-6}M) and for two preincubation periods (1 and 4 hrs). Arsenic was reported to inhibit the level of UVC-induced UDS, although there were no dose- or time-related effects. The actual numbers were not reported in the paper.

Rossman (1983) examined the effect of arsenite on UVC-induced HGPRT mutations in V79 cells. Although no data were provided in this report, the outcome was reported to be negative.

6.4.3 Mammalian Cells In Vivo

The interaction of arsenic and ethylmethanesulfonate (EMS) on levels of chromosome aberrations in vivo was examined in bone marrow and spermatogonial cells of Swiss-albino mice (Poma et al., 1981a). The arsenic dose was 12 mg As/kg combined with either pre- or post-treatment with 200 mg/kg of EMS. No interactions were reported. It should be cautioned that this information has only been reported in abstract form.

Sram (1976) examined the effects of including sodium arsenite in drinking water (7.7×10^{-5}, 7.7×10^{-4} M) on frequencies of tris (1-aziridinyl) phosphine oxide (TEPA)-induced (2 mg/kg) chromosomal aberrations in ICR mouse bone marrow cells. Animals were given arsenic-laced water for 8 weeks, were injected with TEPA, and killed after 24 hours. A total of 250 cells per dose group (5 animals per dose) were scored. Sodium arsenite alone did not appear to induce aberrations, although the spontaneous rate was not clearly indicated. The lower dose of arsenic in drinking water did not affect the frequency of TEPA-induced aberrations. The higher concentration of arsenic resulted in a synergistic increase in the aberration rate. This study was, unfortunately, lacking in experimental details.

Sram (1976) did essentially the same study for the examination of dominant lethality in ICR mice. Animals were maintained for 4 generations prior to mating on arsenic-laced drinking water (7.7×10^{-5}, 7.7×10^{-4} M). TEPA was again given as a single dose of 2 mg/kg. Each treated animal and a control group of 20 males was mated with 40 females per week. The matings continued for 3 to 8 weeks after mutagen application. Sodium arsenite alone did not appear to induce dominant lethals, although the spontaneous rate was not clearly indicated. The lower dose of arsenic in drinking water did not affect the frequency of TEPA-induced dominant lethals. The higher concentration of arsenic resulted in a synergistic increase in the dominant lethal rate. It was not indicated at what time after TEPA administration the increase in dominant lethals occurred; thus, it is not known which germ cell stage was affected. This study also failed to provide experimental details.

6.5 POSSIBLE MECHANISMS OF ACTION

Arsenic is unusual in several respects. First, unlike the overwhelming majority of clastogenic agents, arsenic does not appear to directly damage DNA. Rather, it seems to have its effect through some interference with DNA synthesis. This contention is supported by the observations that arsenic induces chromosomal aberrations and SCE only when it is present during DNA replication. Incubation and removal of arsenic before DNA synthesis has no effect (Nordenson et al., 1981; Crossen, 1983).

Second, arsenic is highly unusual in that it induces chromosomal aberrations and SCE while it fails to induce gene mutations. In this regard, it is like diethylstibesterol, which is another highly unusual carcinogen. X-irradiation, although capable of producing chromosome aberrations as well as gene mutations, is much more potent for the former endpoint. There is a small possibility, however, that the discrepancy for arsenic may be founded on an artifact. Protocols for gene mutation assays generally involve cellular incubation with the test agent for relatively short time periods (2 to 3 hours), while protocols for aberrations often involve the presence of the test agent for one or two entire cell cycles (12 to 48 hours). Thus, in the latter protocol, arsenic would be present for at least an entire S phase for all cells, whereas, when tested for gene mutations, arsenic would be present for only a small fraction of the S phase in approximately one-third to one-half of the cells. Since the evidence available suggests that arsenic has its effect only during DNA replication, this may account for the discrepancy.

Arsenic has long been known to be a sulfhydryl reagent capable of inhibiting a number of thiol-dependent enzyme systems, trivalent forms being much more potent than pentavalent forms (Leonard and Lauwerys, 1980). Thus, one possible mechanism of action for arsenic would be the inhibition of DNA repair enzymes. The work of Rossman (1981) in bacteria and Jung (1969) in human skin cells *in vitro* lends support to this hypothesis. Also, the observations of Sram (1976) on the interactions of arsenic with TEPA for the induction of chromosomal aberrations and dominant lethals support such a contention. The potencies of trivalent and pentavalent arsenicals as sulfhydryl reagents are similar to their potencies as clastogens and SCE-inducing agents. Observations which counter this hypothesis are the reports by Rossman that arsenic has no effect on the frequency of UVC-induced mutations in mammalian cells *in vitro* and the fact that arsenic does not affect the frequency of EMS-induced aberrations *in vivo* (Poma et al., 1981a).

Another possible mechanism for the action of arsenic may be through its occasional incorporation into the DNA backbone in place of phosphorus. There are several lines of evidence to support this mechanism. First, for this to occur, arsenic would have to be present during DNA synthesis and would have no effect on non-dividing cells. Second, such a mechanism could explain why arsenic is clastogenic (such a bond would be weaker than the normal phosphodiester bond) but does not induce gene mutation. Third, arsenic has been shown to cause strand breaks in DNA (Fornace and Little, 1979). X-irradiation, a potent clastogen and poor inducer of gene mutations, also predominantly causes strand breaks as its major DNA lesion. An argument against such a mechanism is the observation that the trivalent forms are more potent than pentavalent forms, while pentavalent arsenic should be more likely to substitute for phosphorus in DNA. Therefore at present, there is no single, unambiguous explanation for the mechanisms by which arsenic breaks chromosomes or induces SCE.

7. ARSENIC CARCINOGENICITY

The case for the association of inorganic arsenic with skin and lung cancer as well as other visceral carcinomas has been extensively reviewed (Arsenic NAS, 1977; IARC, 1973 and 1980; NIOSH, 1975; Hernberg, 1977; Sunderman, 1976, Pelfrene, 1976; Kraybill, 1978; Wildenberg, 1978; Pershagen and Vahter, 1979; WHO, 1981). The following chapter is divided into three major sections discussing human studies, animal studies and quantitative carcinogenic risk estimates.

7.1 HUMAN STUDIES

The literature on arsenic carcinogenicity in humans is summarized in Table 7-1. This subsection on human studies will first focus on clinical pathophysiological aspects of arsenic carcinogenesis, followed by pertinent epidemiological studies of arsenic-induced carcinogenesis.

7.1.1 Clinical Aspects of Human Arsenic Carcinogenesis

In man, chronic exposure to arsenic induces a characteristic sequence of changes in skin epithelium, proceeding from hyperpigmentation to hyperkeratosis which may be histologically described as showing keratin proliferation of a verrucose nature with derangement of the squamous portions of the epithelium or may even be described in some cases as squamous cell carcinomas.

Late onset skin cancers, associated with arsenic exposure, appear to be of two histopathological types: squamous carcinomas in the keratotic areas and basal cell carcinomas. In one study dealing with skin cancer after prolonged use of Fowler's solution (Neubauer, 1947), the ratio of types was approximately 1:1.

Arsenic-associated skin cancers differ from those of ultraviolet light etiology by occurring on areas generally not exposed to sunlight, e.g. palms and soles, and occurring as multiple lesions (Arsenic. NAS, 1977; Pershagen

TABLE 7-1. SUMMARY OF CASE REPORTS AND EPIDEMIOLOGIC STUDIES OF CANCER OR PRECANCEROUS LESIONS IN PERSONS EXPOSED TO ARSENIC

Media	Study Population	Author(s)	Type of Study	Results	Highlights and Deficiencies
Air	Smelter workers-Tacoma, Washington (Analysis of deaths for 1946-1960)	Pinto and Bennett (1963)	Proportionate mortality	No difference in lung cancer proportionate mortality between exposed and unexposed workers.	Workers leaving the plant before retirement were not included. In the classification of workers by exposure, the "non-exposed" group apparently were exposed since they also had high levels of arsenic in the urine.
	Smelter workers-Tacoma, Washington (follow-up from 1950-1971)	Milham and Strong (1974)	Cohort	40 observed lung cancer deaths versus 18 expected ($P < 0.001$).	Urinary arsenic levels of persons living around the smelter decreased with distance from the smelter.
	Smelter workers-Tacoma, Washington (follow-up from 1949-1973)	Pinto et al. (1977)	Cohort	32 observed respiratory cancer deaths, versus 10.5 expected ($P < 0.05$). Dose response seen by urinary arsenic levels and by duration and intensity of exposure.	Study consisted of only pensioners.
	Smelter workers-Tacoma, Washington (Follow-up from 1941-1976)	Enterline and Marsh (1982)	Cohort	104 respiratory cancer deaths observed versus 52.5 expected ($P < 0.01$). Dose response found by intensity and duration of exposure.	Short-term high-intensity arsenic exposures appeared to have a greater effect than did long-term low-intensity exposures; SO_2 exposure was found to have little or no effect.
	Smelter workers-Anaconda, Montana (Follow-up from 1938 to 1963)	Lee and Fraumeni (1969)	Cohort	147 respiratory cancer deaths observed versus 44.7 expected ($P < 0.01$). Dose response found by intensity and duration of exposure.	A dose response was found between exposure to sulfur dioxide and respiratory cancer mortality. Exposure to sulfur dioxide could not be separated from exposure to arsenic, however.
	Smelter workers-Anaconda, Montana (Follow-up from 1964 to 1977)	Lubin et al. (1981)	Cohort	146 respiratory cancer deaths versus 88.7 expected ($P < 0.01$).	Exposure to sulfur dioxide was not found to have an independent effect on cancer risk.
	Sample of 1800 of the smelter workers-Anaconda, Montana (Follow-up from 1938-1977)	Welch et al. (1982); Higgins et al. (1982)	Cohort	24 respiratory cancer deaths versus 4.6 expected ($P < 0.01$) in the heavy exposure category. Dose response found by intensity (both time-weighted average and ceiling level categories) of exposure.	Analysis of lung cancer mortality by SO_2 exposure found that SO_2 did not play an important role in the respiratory cancer process.

TABLE 7-1. (continued)

Media	Study Population	Author(s)	Type of Study	Results	Highlights and Deficiencies
Air	Smelter workers-Anaconda, Montana (Follow-up from 1938 to 1977)	Lee-Feldstein (1983)	Cohort	302 respiratory cancer deaths observed versus 105.8 expected ($P < 0.01$). Dose response found by duration and exposure.	
	Lung cancer from the parish where the Ronnskar smelter is located	Axelson et al. (1978)	Case-control	For smelter workers, the lung cancer mortality odds ratio was 4.6; there also was a significantly ($P < 0.02$) elevated risk of leukemia and myeloma among smelter workers.	Exposure to sulfur dioxide did not appear to be associated with lung cancer.
	Lung cancer deaths in the city of Saganoseki-machi, Japan	Kuratsune et al. (1974)	Case-control	58% of lung cancer cases were found to be former smelter workers versus 15.8% in controls.	The cause of death listed on the death certificate was validated using detailed pathologic analysis.
	Copper smelters in Sagnoseki-machi, Japan	Tokudome and Kuratsune (1976)	Cohort	29 trachea, lung, and bronchus cancer deaths versus 2.44 expected ($P < 0.01$); 3 observed colon cancer deaths versus 0.59 expected ($P < 0.05$). A lung cancer dose response was seen by length of employment and level of exposure.	The latent period ranged from 13 to 50 years, with an average of 37.6 years.
	Smelter workers in Magna, Utah	Rencher et al. (1977)	Proportionate mortality and cohort	7 percent of the deaths were lung cancer deaths compared to 0 to 2.2 percent for other factory workers and 2.7 percent for the State; the lung cancer death rate was found to be 10.1 per 10,000 versus 2.1 and 3.3 per 10,000 for mine workers and the State, respectively.	
	Residents living near a smelter in El Paso. Texas	Rom et al. (1982)	Case-control	No association was found between lung cancer and distance from the plant.	Effects of migration, smoking and occupation were not considered.

TABLE 7-1. (continued)

Media	Study Population	Author(s)	Type of Study	Results	Highlights and Deficiencies
Air	Residents of Deer Lodge and Silver Bow Counties, Montana	Newman et al. (1976)	Ecological correlation	There was an increase found in the incidence of lung cancer among men. In one of the cities there was also an increase in lung cancer among women.	No adjustment was made for cancer cases which may be occupational.
	All counties in the United States with smelters	Blot and Fraumeni (1975)	Ecological correlation	Average lung cancer mortality rates were significantly elevated for both males and females in 36 counties with smelters processing copper, lead, or zinc ores.	
	Residents near a smelter in Utah	Lyon et al. (1977)	Case-control	No association between cancer and distance from the smelter was found.	Lymphoma cases which may have an association with arsenic exposure were used as controls. Effects of smoking, migration, and occupation were not considered.
	Residents near Ronnskarverken smelter in northern Sweden	Pershagen et al. (1977)	Ecological correlation	A significantly higher mortality rate for lung cancer was noted for men in the exposed area. The increase was no longer significant when occupational cases were excluded, however.	When excluding occupational cases of lung cancer from the study population, lung cancer cases for a comparable occupational group were not excluded from the comparison population.
	Population surrounding an arsenical pesticide facility	Matanoski et al. (1976, 1981)	Ecological correlation	The lung cancer mortality for males in the census tract in which the plant was located was 3-4 times higher than the control tracts ($P < 0.05$).	The difference in the lung cancer mortality rate in the index tract could not be explained by occupation.
	Arsenical sheep dip manufacturing workers	Hill and Faning (1948)	Proportionate mortality	29.3 percent of deaths were due to lung cancer versus 12.9 percent of deaths among workers in the same geographic area who were not exposed to arsenic ($P < 0.05$). The excess in cancer deaths was mainly due to an excess in lung cancer and skin cancer.	

TABLE 7-1. (continued)

Media	Study Population	Author(s)	Type of Study	Results	Highlights and Deficiencies
Air	Arsenical pesticide manufacturing workers	Ott et al. (1974)	Proportionate mortality and cohort	16.2% and 3.5% of deaths in the exposed group were from cancer of the respiratory system and from lymphatic and hematopoietic cancers, except leukemia, respectively, versus 5.7 and 1.4% in the controls; the cohort mortality study found 20 respiratory cancer deaths and 5 deaths of the lymphatic and hematopoietic tissues versus 5.8 and 1.3 expected, respectively (both significant at $P < 0.01$).	A respiratory cancer mortality dose response was not found below an average dosage of 3890 mg of arsenic, but above that dosage there was a good dose response. It should be noted, however, that all of the respiratory cancer at or below 3890 mg had less than one year of exposure. Thus, it is unlikely that those deaths were due to arsenic exposure.
	Retirees of an arsenical pesticide plant in Baltimore, Maryland (follow-up from 1960 to 1972)	Baetjer et al. (1975)	Proportionate mortality and cohort	The proportionate mortality ratio (PMR) was 6.58 for respiratory cancer ($P < 0.05$); for cause-specific mortality the observed-to-expected ratios were 16.67 for respiratory cancer and 50 for lymphatic cancer (both with $P < 0.05$).	The cohort study was limited to pensioners only.
	Retirees of an arsenical pesticide plant in Baltimore Maryland (follow-up 1946 to 1977)	Mabuchi et al. (1979)	Cohort	12 observed lung cancer deaths versus 3.6 expected ($P < 0.05$); a dose response by duration of employment was seen for those with exposure of high intensity.	
	Wenatchee Valley orchard workers in the state of Washington	Nelson et al. (1973)	Cohort	No difference was found between the cohort and the state of Washington for overall cancer mortality or for lung cancer mortality.	
	German vintners	Roth (1958)	Proportionate mortality	Of 47 autopsies among vintners with chronic arsenic intoxication, 64% of the deaths were due to cancer, 60% to lung cancer; 6 of the 47 and 13 of the 47 were reported to have liver and skin tumors, respectively.	No controls were used. There was no indication of how the autopsy cases were selected.

TABLE 7-1. (continued)

Media	Study Population	Author(s)	Type of Study	Results	Highlights and Deficiencies
Water	Residents of a section of Taiwan with high levels of arsenic in the drinking water	Tseng et al. (1968); Tseng (1977)	Cross-sectional	A skin cancer prevalence rate of 10.6/1000 for those drinking well water was found compared to 0/1000 for a control area. The skin cancer rate followed a dose response by arsenic concentration in the water.	
	Sample of inhabitants of Antofagasta, Chile	Borgono and Greiber (1972)	Cross-sectional	Abnormal skin pigmentation and hyperkeratosis were found in Antofagasta where high arsenic levels in the water were reported. No skin cancers were reported, however.	
	Residents of certain departments in the province of Cordoba, Argentina, exposed to high levels of arsenic in the water	Bergoglio (1964)	Proportionate mortality	The proportion of cancer deaths was higher than for the province as a whole (23.84% versus 15.3%, $P < 0.05$). Of the cancer deaths, respiratory cancer was reported to be 35% and skin cancer 2.3%.	The data were apparently not age-adjusted. No comparison was made of the percentage of respiratory and skin cancer deaths in the affected departments with the respective proportion for Cordoba Province.
	Patients at a dermatology clinic in eastern Argentina	Arguello et al. (1938)	Case reports	The largest proportion of cases in the clinic (82%) came from areas with the highest incidence of chronic endemic regional arsenical intoxication.	The study was not population-based.
	Residents in an area of Lane County, Oregon	Morton et al. (1976)	Cross-sectional	The prevalence of skin cancer was not found to be associated with arsenic concentrations in the water.	The sample size was relatively small.
	Residents of Hinckley and Deseret, Utah, where relatively high arsenic contamination of the drinking water supplies existed and Delta, Utah, which had very little arsenic contamination of the water supply	Southwick et al. (1981)	Cross-sectional; mortality comparison	There was no dermatological evidence of arsenic ingestion in Hinckley or Deseret. No evidence of unusual cancer mortality patterns in Hinckley was found.	Sample size was very small-only 145 in the exposed group, 105 in the controls. The town of Hinckley is rather small for a mortality analysis. The population was only 400 in 1970.

TABLE 7-1. (continued)

Media	Study Population	Author(s)	Type of Study	Results	Highlights and Deficiencies
Water	353 cases of Blackfoot disease and 353 controls matched for sex and age in an area of Taiwan with an endemic arsenic contamination of the water supply	Ch'i and Blackwell (1968)	Case-control	Significantly ($P < 0.01$) more cases than controls were found to consume deep well water (known to be contaminated with arsenic) and population had a low socioeconomic status.	
	Two persons living in an area of Taiwan with endemic high levels of arsenic in the water supply	Astrup (1968)	Case report	Both individuals had developed Blackfoot disease.	
Water, soil, dust	One female child	Falk et al. (1981b)	Case report	Child, with a history of pica, developed hepatic angiosarcoma with associated congenital dysplasia of the liver and an infantile benign hemangioendothelioma suggesting pre-natal as well as post-natal etiological factors.	
Milk	A group of children in Japan	Yamashita et al. (1972)	Cohort	Hyperpigmentation and depigmentation were found to be prevalent in about 15 percent of the survivors approximately 15 years after exposure.	There was no comparison group.
Arsenical medicinals	Patients being treated with arsenical medicinals	Hutchinson (1888)	Case reports	Six patients treated with arsenical medicinals exhibited the keratotic lesions associated with arsenical poisoning.	
Arsenical medicinals	Patients being treated for skin diseases and various internal disorders with arsenical medicinals.	Neubauer (1947)	Case reports	143 individuals with epitheliomas and who had taken arsenical medicinals.	

TABLE 7-1. (continued)

Media	Study Population	Author(s)	Type of Study	Results	Highlights and Deficiencies
Arsenical medicinal	Patients treated with Fowler's solution (an arsenical medicinal) by a private practitioner	Fierz (1965)	Cohort	106 of the 262 patients reporting for physical examination reported hyperkeratosis; 21 cases of skin cancer were also found. The response increased with increasing dose.	Less than 45% of the cohort presented themselves for physical examination, and the author himself reported that the patients reporting for examination were not a representative sample. Furthermore, no controls were used.
Arsenical medicinal	Patients treated with arsenical medicinals	Reymann et al. (1978)	Cohort	Of 389 persons treated with arsenical medicinals, 41 internal malignant neoplasms were found to occur versus 44.6 expected. No increase in internal malignant neoplasms was found by dose.	There was no control group. Internal organs presumably include the lung, and there was no control for smoking, a major confounder with regard to lung cancer.
Arsenical medicinal (Fowler's Solution)	Patients treated with Fowler's solution	Cuzick et al. (1982)	Cohort	A statistically significant association of ingestion of Fowler's solution with deaths from internal malignancies was not found. A subset of 142 of the 487 in the cohort was examined and 49% were found to show dermal signs of arsenicism including skin cancer (11%).	In the subset of 142, deaths from internal malignancies occurred only in those who had previously demonstrated dermal signs of arsenicism, leading the authors to conclude that perhaps persons who show signs of arsenicism are at a greater risk of death from internal malignancies.
Arsenical medicinal (Fowler's Solution)	Male patient	Regelson et al. (1968)	Case report	Hemangioendothelial sarcoma of the liver.	
Arsenical medicinals (Fowler's Solution)	27 cases exposed to arsenic either via arsenical drugs or via occupation	Sommers and McManus (1953)	Case reports	Skin carcinomas were reported in all cases; visceral cancers were reported for some of the cases.	
Arsenical medicinal	Male patient	Nurse (1978)	Case report	Adenocarcinoma of the kidney	Patient treated with a variety of other drugs before developing kidney cancer.

TABLE 7-1. (continued)

Media	Study Population	Author(s)	Type of Study	Results	Highlights and Deficiencies
Arsenical medicinal	2 male patients 1 female patient	Knoth (1966)	Case reports	Female developed mammary carcinoma and skin cancer; 1 male patient developed a reticulosarcoma of the glans penis; 1 male developed skin cancer.	
Arsenical medicinals, occupational exposure, and air (occupational) exposure	16 vintners who were arsenical pesticide users; patient who took arsenical medicinals	Braun (1958)	Case reports	Skin and visceral cancers were reported among the vintners; skin and lung cancers were reported for one patient taking an arsenical medicinal; skin cancer was reported for the other patient who took an arsenical medicinal.	
Arsenical medicinals (Fowler's Solution)	2 male patients	Morris et al. (1974)	Case reports	One patient developed skin pigmentation, skin tumors, carcinoma of the larynx, and a probable bronchial carcinoma; the other developed skin pigmentation and keratosis. Both developed noncirrhotic portal hypertension.	
Arsenical medicinal (Fowler's Solution)	Female patient	Prystowsky (1978)	Case report	Woman developed nasopharyngeal cancer; also developed palmar and plantar keratosis.	
Arsenical medicinals (Fowler's (Solution)	4 male patients and 1 female patient	Popper et al. (1978)	Case reports	The cases developed angiosarcoma of the liver.	
Arsenical medicinal (Fowler's Solution)	1 male patient	Lander et al. (1975)	Case report	The patient developed angiosarcoma.	

TABLE 7-1. (continued)

Media	Study Population	Author(s)	Type of Study	Results	Highlights and Deficiencies
Arsenical medicinal (Fowler's Solution)	1 male patient	Roat et al. (1982)	Case report	The patient developed both angiosarcoma and skin cancer.	
Arsenical medicinal (Fowler's Solution)	7 male patients and 1 female patient	Falk et al. (1981a)	Case reports	The cases developed angiosarcoma.	
Wood preservative	Wood treaters in Hawaii	Gilbert et al. (1983)	Cross-sectional and cohort	No association of arsenic exposure with cancer mortality or morbidity was found.	The sample size was small (only 88 in the study cohort and 61 in the controls in the cross-sectional study; 125 in the mortality study; 182 in the morbidity study).

and Vahter, 1979; WHO, 1981; Tseng, 1977; Sunderman, 1976). This appears to be the case for medicinal (Neubauer, 1947), environmental (Tseng et al., 1968), and occupational (Roth, 1958; Braun, 1958) exposure. The time lag between initiation of exposure and occurrence of skin cancer has been reported to range from 6 to 50 years for arsenical medicinally-induced skin cancer. The minimal latency period for skin cancer in the most reliable epidemiologic study of arsenic-contaminated drinking water was reported to be 24 years.

The amount of data on the histological classification of lung tumors associated with occupational arsenic exposure is limited. Newman et al. (1976) report that arsenic-associated lung cancers are usually the poorly differentiated type of epidermoid bronchogenic carcinoma. These investigators studied worker groups with diagnosed lung cancer in copper-mining and smelting communities in Montana. Of 25 smelter workers, 4 had well-differentiated epidermoid carcinoma, 10 poorly differentiated epidermoid carcinoma, 7 small-cell undifferentiated epidermoid carcinoma and 3 acinar-type adenocarcinoma. Copper miners and "non-copper" control individuals had lung cancer profiles which were similar to each other.

The time period between initiation of exposure and the occurrence of arsenic-associated lung cancer was found in a couple of studies to be on the order of 35-45 years (Lee and Fraumeni, 1969). Recently, a latency period of <20 years was reported by Enterline and Marsh (1980; 1982) based upon their studies of copper smelter workers in Tacoma, Washington. Tokudome and Kuratsune (1976) found that the latent period for lung cancer ranged from 13 to 50 years.

The association of other visceral cancers with arsenic exposure has been noted in a number of reports and has been reviewed elsewhere (WHO, 1981; Arsenic. NAS, 1977; NIOSH, 1976; IARC, 1973). For example, hemangiosarcoma of the liver, a rare form of cancer, has been diagnosed in workers exposed to arsenic and in non-occupationally arsenic-exposed individuals (Roth, 1958; Regelson et al., 1968; Lander et al., 1975; Falk et al., 1981a and b; Roat et al., 1982). Morris et al. (1974) have postulated that the peculiar hepatic fibrosis associated with arsenic-induced portal hypertension is a precursor state for subsequent progression to hepatic angiosarcoma. Popper et al. (1978) have noted that the hepatic fibrosis and hypertension seen in humans with Thorotrast, vinyl chloride or arsenic exposure are also induced by agents which presumably also have a role in hepatic angiosarcoma.

Other cancers noted in arsenic-exposed subjects include: lymphomas and leukemia (NIOSH, 1976; Ott et al., 1974); renal adenocarcinoma (Sommers and McManus, 1953; Nurse, 1978); and nasopharyngeal carcinoma (Prystowsky et al., 1978).

Pelfrene (1976) has criticized the reports of internal malignant neoplasms associated with arsenic exposure on the basis of the relative rarity of their detection in large-scale studies of chronic arsenic exposure such as that of Tseng (1968, 1977). More recently, Reymann et al. (1978) reported on a study of a group of 389 patients who took arsenic medicinally in the 1930s. An excess of internal cancers was not observed in the total cohort when compared with the expected incidence of malignant internal neoplasms based on the Danish Cancer Registry. The size of the cohort was probably too small to have detected an increase, however.

7.1.2 Epidemiological Aspects of Human Arsenic Carcinogenesis

7.1.2.1 Cancer of the Lung--A large number of reports are available on possible associations between occupational exposure to arsenic and cancer of the respiratory system. As is common in studies of this type, exposure data are very uncertain and the arsenic exposure is not always clearly defined regarding the physicochemical properties of the arsenic compounds. The picture is often confused by simultaneous exposure to other agents, especially sulfur dioxide and metals. Data on smoking are often lacking or incomplete.

An excess mortality in respiratory cancer has especially been noted among workers engaged in the production and usage of pesticides, and among smelter workers.

In 1948, Hill and Faning presented data on proportional mortality rates among British workers exposed to a mixture of ingredients--including sodium arsenite, powdered sulfur and soda ash--used in the manufacture of a sheep-dip powder. Between 1910 and 1943, 75 deaths had occurred among workers in the sodium arsenite factory and 1,216 deaths had occurred among workers in the same area but without known exposure to arsenic. Proportionate mortality analysis showed that of the deaths among factory workers, 29.3 percent had died from cancer, whereas the corresponding figure for the other workers was 12.9 percent. The excess in cancer deaths among the factory workers was mainly due to an excess in lung cancer, 31.8 percent of all cancer deaths compared with 15.9 percent; and in skin cancer, 13.6 percent compared with 1.3 percent.

Among the factory workers, chemical workers, who were the workers most closely associated with arsenite production, had a higher proportion of cancer deaths than did the factory workers as a group. Furthermore, all lung cancer deaths had occurred among the chemical workers.

Arsenic in the air of the sodium arsenite factory was determined in 1945-46 (Perry et al., 1948) and concentrations up to 4 mg As/m^3 were found by sampling for 10 minutes. No data were given on the age of the deceased, and smoking habits were not recorded. The data do not allow any conclusions about exposure before 1943. Nevertheless, this study indicated that there might be an increased risk for respiratory cancer in the manufacture of arsenic-containing pesticides, and studies in two United States plants have given further support.

Ott et al. (1974) studied the mortality of workers in one of these two chemical plants. From 1919 to 1956 one unit formulated and packaged insecticides containing arsenic in the form of lead arsenate, calcium arsenate, copper acetoarsenite, and magnesium arsenate. During this period, the proportions of the different compounds varied. The main product was lead arsenate. The size of the workforce was about 30 in 1928 and 100 in 1948. Turnover was high with less than 25 percent of the men remaining with the unit for more than one year. Arsenic concentrations in air in 1943 were between 0.18 and 19 mg As/m^3 in the packaging area; in 1952 concentrations were 1.7-40.8 mg As/m^3 and 0.26-7.5 mg/m^3 in the drum dryer area and packaging area, respectively. By combining job classifications and air arsenic data, four exposure classifications were obtained with estimated arsenic exposures (8-hr TWA) of 5, 3, 1, and 0.1 mg/m^3. The total dosage was then calculated for each individual by multiplying air levels with the number of days at work and assuming that 4 m^3 were inhaled during a working day.

Mortality was studied by analysis of proportionate mortality and by a retrospective cohort analysis. Nearly 2,000 employees in the factory had died between 1940 and 1972. One hundred seventy-three were identified as having worked one or more days in the arsenical production unit and who then either worked for the company until death or died after retirement.

Ott et al. (1974), after adjusting for age and year of death, compared the differences in proportionate mortality between the study group and the controls. Among the exposed, respiratory cancer accounted for 16.2 percent of the deaths compared with 5.7 percent in the controls ($p < 0.001$). There was also a significant increase ($p < 0.05$) in deaths from lymphatic and hematopoietic cancers, except for leukemia. Table 7-2 shows the observed-to-expected ratios for respiratory cancer in relation to exposure. (In the original table, dosage estimates were based on 4 m^3 inhaled per 8 hr and expressed as the natural logarithms.

TABLE 7-2. OBSERVED AND EXPECTED DEATHS DUE TO RESPIRATORY MALIGNANCIES, BY EXPOSURE CATEGORY

TWA Concentration x months of exposure	Average dosage mg	No. of total deaths†	Respiratory Malignancy Deaths		
			Observed*	Expected	Ratio Observed/Expected
<1	105	26	1 (1)	1.77	.6
1-1.9	316	17	2 (2)	1.01	2.0
2-3.9	630	24	4 (4)	1.38	2.9
4-5.9	1050	22	3 (3)	1.36	2.2
6-11.9	1991	27	3 (3)	1.70	1.8
12-23.9	3890	18	2 (2)	.97	2.1
24-59.9	8833	13	3 (1)	.77	3.9
60-95.9	16257	13	5 (0)	.79	6.3
96+	74332	13	5 (0)	.72	7.0

†n = 173, 138 workers had less than one year of exposure.

*Number in brackets shows respiratory cancer deaths in workers exposed less than one year.

Source: Modified from Ott et al. (1974), and Blejer and Wagner (1976).

In the present table, dosage is expressed in mg and is based on 10 m^3 inhaled air.) There is no tendency towards a dose-response relationship at total exposures from 105 to 3890 mg, but a sharp increase is noted at higher dosages. Furthermore, Blejer and Wagner (1976) reported that of the 173 deaths, 138 had occurred among workers with less than one year of exposure. In that group, 16 deaths were due to respiratory cancer. As seen in Table 7-2, 15 of those deaths occurred in the six groups with average total exposures estimated to be from 105 to 3890 mg. There are no quantitative data on other compounds that these workers might have been exposed to during their total time with the company; however, it is known that in addition to the arsenic-containing insecticides, the plant processed and packaged several other products, the most important of which were powdered sulfur and dry lime sulfur.

The retrospective cohort analysis was based on a roster of 603 workers who had worked for at least one month in the actual unit between 1940 and 1973. Workers leaving the company before retirement as well as workers who had been exposed to asbestos were not included in the analysis. It was stated that virtually all men with at least one year of exposure had been identified. Person-years, by 10-year age groups, for five calendar year groups were calculated and expected number of deaths were calculated by using United States white male mortality data. Table 7-3 shows that by this analysis a significant increase ($p < 0.01$) in deaths due to respiratory cancer was found among exposed workers. There was also a significant increase ($p < 0.01$) in the number of deaths due to malignant neoplasms in lymphatic and hematopoietic tissues, except leukemia.

A chemical plant in Baltimore was the subject of two studies concerning the working environment and one study concerning the outer environment. This plant started producing arsenic acid in the early 1900s and in the early

TABLE 7-3. OBSERVED AND EXPECTED DEATHS FOR SELECTED CAUSES IN RETROSPECTIVE COHORT ANALYSIS (1940-1973)

	Observed deaths	Expected deaths (U.S. white males)	Ratio of observed to expected deaths
All causes	95	113.5	.84
Malignant neoplasms, total	35	19.4	1.80
Respiratory system	20	5.8	3.45†
Digestive organs & peritoneum	7	6.3	1.11
Lymphatic & hematopoietic tissues except leukemia	5	1.3	3.85†
All other sites	3	6.0	.50
Diseases of cardiovascular system	41	58.5	.70
Emphysema, chronic bronchitis, & asthma	4	2.8	1.43
All external causes	9	12.7	.71
All other causes	6	20.1	.30

† $p < 0.01$

Source: Ott et al. (1974).

1950s production of arsenical pesticides started. Lead arsenate, calcium arsenate and sodium arsenate were among the compounds produced. Production terminated in 1974 (Matanoski et al., 1976). The plant also produced chlorinated hydrocarbons and organic phosphates. Data on air concentrations of arsenic are lacking.

In the first study, Baetjer et al. (1975) reported on both the proportionate mortality and age-specific death rates of workers who had retired between 1960 and 1972 and generally had at least 15 years' employment. Seventeen of 22 deaths among male white workers and two of five among female workers were due to malignant neoplasms. The female deaths did not appear to be related to occupational exposure, whereas, the male deaths did. Of the 17 malignant neoplasms in males, 10 were in the respiratory tract, 3 were lymphosarcomas, and the remaining 4 were of other tissues. The proportionate mortality analysis, based on mortality data for the city of Baltimore, showed an observed/expected ratio of 6.58 for respiratory cancer ($p < .05$) and 15.79 for cancer of the lymphatic and hematopoietic system ($0.1 < p < 0.05$). The age-specific death rate analysis showed that deaths from respiratory cancer were 16.67 times the expected, and, that for lymphosarcomas, the observed number was 50 times that of the expected number. The authors reported that observed and expected rates for non-cancer deaths did not differ significantly ($p < 0.05$).

In a second study (Mabuchi et al., 1979), a follow-up was made of workers employed from 1946 to 1974. Since exposure data were lacking, an attempt was made to classify workers according to exposure to arsenicals and non-arsenicals. A roster of 3,141 workers was obtained. Since 2,189 workers had been employed for less than 4 months, a 20 percent random sample was drawn from that population and together with the remaining 952 workers constituted the study population: 1,050 males and 343 females, mainly white. Exposure assessments were made, and the workers were categorized into one of six exposure groups.

Of the study population, 240 had died: 197 males and 43 females. Expected deaths were calculated from the city of Baltimore statistics. The observed/expected ratios for lung cancer were analyzed by exposure, year of first employment, and duration of employment. A statistically significant increase in lung cancer mortality (SMR = 336, $p < 0.05$) was found among "predominantly arsenical production" workers. There was a clear lung cancer mortality dose response among these workers by duration of exposure. Those employed 15-24 years and 25+ years both had statistically significant ($p < 0.05$) lung cancer SMRs (1365 and 2750, respectively). Interestingly, statistically significantly elevated SMRs were only found in "predominantly arsenical production" workers, but not in workers engaged entirely in arsenical production. Further analysis revealed that the proportion of workers engaged entirely in arsenical production for 5 years or more was relatively low (1 percent), while the proportion of workers exposed predominantly, but not entirely, to arsenic for 5 years or more was much higher (29 percent). This difference in duration of exposure may have accounted for the absence of excess lung cancer mortality among the workers engaged entirely in arsenical production. Data on smoking were not obtained.

Occupational exposure to arsenical pesticides has been common among vintners and agricultural workers. Exposure has mainly been to lead arsenate. In a study of orchard workers who used lead arsenate in the Wenatchee Valley in the state of Washington, Nelson et al. (1973) found a deficit of lung cancer mortality for the period 1938-68 when compared with mortality for the state of Washington.

Because the results of this study were at variance with previous evidence on the long-term effects of arsenic exposure, NIOSH reviewed data from other sources and used alternative procedures in an attempt to verify the findings. Analysis of respiratory cancer mortality data by occupational category for the state of Washington for the period 1950-71 found that orchardists had 19 percent more deaths from respiratory cancer than expected. An analysis of cancer mortality rates for 1950-69 for the counties comprising the locale from which the orchardist sample was drawn by Nelson et al. found that males had a 7 percent higher respiratory cancer rate than expected among males. The county from which most of the orchardists were drawn had a 31 percent excess ($P < 0.01$) of lung cancer mortality. Thus, it was NIOSH's (1975) conclusion that the Nelson et al. report did not accurately depict the cancer experience of persons exposed to lead arsenate spray in the Wenatchee Valley.

Several studies in Germany indicate that workers exposed to arsenic trioxide when spraying vineyards had a high mortality in cancer, especially lung cancer. In one report (Roth, 1958), it was stated that of 47 autopsies among vintners with chronic arsenic intoxication, 30 (64 percent) were due to cancer, and 18 to lung cancer (60 percent of all cancer deaths). The author did not state how the cases were selected, nor were controls used in the study.

Gilbert et al. (1983) studied a group of 182 workers in Hawaii exposed for at least 3 months during the period 1960-1981 to the wood-treating chemicals chromated-copper-arsenate (CCA), pentachlorophenol (penta), tributyl tin oxide (TBTO) and lindane. The study was divided into two parts: (1) a cohort comparison study of 88 workers and 61 controls and (2) a historic prospective study which consisted of a morbidity and a mortality analysis of the entire cohort.

In the cohort comparison study, 61 controls were matched on the basis of age, sex, race, level of physical activity, and weight to 88 wood treatment workers who were "qualified and agreed to participate in the study." Controls were recruited from among the membership of carpenters', ironworkers', masons', plumbers', and stevedores' unions and from the names of friends and relatives referred to the study by participating members of the occupational cohort. Fourteen of the controls were reported to be carpenters and 13 of them had had exposure to either CCA or penta or both. Their urine arsenic levels, however, were reported not to differ significantly from those of other controls. The exposed group and the control group were each given a comprehensive health examination consisting of a questionnaire and clinical and laboratory tests including analysis of a urine sample for penta, arsenic, copper, chromium, and tin levels. Only penta was found to be significantly elevated in the urine of the exposed group over that of the controls. The authors reported that there were no "clinically significant" differences between the exposed group and the controls. No significant differences were found between the wood treaters and controls with respect to educational level, smoking history, or alcohol consumption.

The historical prospective study identified three cases of cancer, one by means of the questionnaire in the cross-sectional study and the other two by means of the Hawaii Tumor Registry. One of the cancer cases was a bladder cancer case; the other two were colorectal cancer cases. For the mortality analysis, the vital status of 125 of the 182 workers (69 percent) was able to be determined. Of these 125, six deaths occurred, five from cardiovascular disease, the other from an unknown cause. The authors calculated that eight deaths would have been expected in this group, three of which would have been from cancer. The authors did not state the basis of the expected number of cancer deaths.

There are limitations in the study design and sample size of this study which make the results with regard to the evaluation of a cancer risk among the wood treaters inconclusive. A cohort comparison study on the basis of a single medical examination is an inappropriate approach to determine if a cancer risk exists in the wood treater group, particularly when the group consists primarily of persons who are currently employed as wood treaters (60 of 88 were current employees). Most persons who have developed cancer will either have died or left the work force. In addition, the sample size (88 current or former wood treaters and 61 controls) would have been too small to detect an excess cancer risk. Also, it should be noted that the inclusion in the controls of 13 carpenters who had been exposed to arsenic-treated wood certainly presents a potential bias, despite the fact that the authors claimed that the levels of arsenic in the urine of these 13 were not significantly different from those in other controls. In this regard it should be noted that no effort had apparently been made to restrict the intake of seafood or other foods or liquids, which might have elevated urine arsenic levels, prior to collection of the urine samples. The mean level of urinary arsenic in both the study group and in the controls was significantly ($P < 0.01$) higher than what the authors reported would be a normal level.

The historic prospective study also had limitations. Again, the sample size was too small to adequately determine whether an excess risk of cancer existed in the study cohort. Only 125 were included in the mortality study and only 182 were included in the morbidity study. Arsenic exposure via inhalation and ingestion is known to be associated with lung and nonmelanoma skin cancer, respectively. Nonmelanoma skin cancer is rarely fatal and thus, an excess risk of skin cancer would not have been detected in such a small mortality study. The Hawaii Tumor Registry, which was one of the primary sources of information used to identify cancer cases for the morbidity study, does not even report nonmelanoma skin cancer.

There is also a question of whether the authors had sufficiently allowed for a cancer latency period in their study. The authors did not indicate the length of follow-up of the cohort members. Some indication is provided, however, in the data reported by the authors on the length of employment of the 88 workers in the study cohort of the cohort comparison study. These workers were part of the population in the incidence and mortality studies. Of the 88 workers, 60 had worked 10 years or less and 80 had worked 15 years or less at the time of the physical examination in 1981. Twenty-two of the study cohort in the cohort comparison study were former employees. At the minimum, assuming that the workers had had no breaks in employment, 58 of 125 (46 percent) in the mortality study would have been followed for only 15 years or less and 38 of 125 (30 percent) would have been followed for only 10 years or less. In the morbidity study, if, again, no breaks in employment of the workers are assumed, 58 of 182 (32 percent) would have been followed for only 15 years or less and 38 of 182 (21 percent) would have been followed for only 10 years or less. In conclusion, this study is inadequate to conclude whether an excess risk of cancer exists in the wood treater population in Hawaii.

Occupational exposure to arsenic also occurs in smelters where exposure is predominantly to arsenic trioxide. Several studies have been done on mortality among workers at the copper smelter in Tacoma, Washington. Pinto and Bennett (1963) reported on the proportionate mortality of 229 workers from 1946 to 1960. Workers leaving the plant before retirement were not included. The proportionate mortality of the smelter workers was compared with that of males in the state of Washington in 1958. Of all cancer deaths, the respective proportions of lung cancer were 41.9 and 23.7 percent, respectively. The authors then classified the smelter workers according to arsenic exposure and did not find any difference between exposed and non-exposed. However, the "non-exposed" had elevated arsenic levels in urine, suggesting possible exposure.

More extensive studies have since been published, showing an increase in lung cancer among arsenic-exposed workers (Milham and Strong, 1974; Pinto et al., 1977; Pinto et al., 1978; Enterline and Marsh, 1980; Enterline and Marsh, 1982).

Milham and Strong (1974) examined county records from 1950 to 1971 to find the number of deaths due to respiratory cancer among county residents employed at the smelter. Expected number of cases were calculated for the smelter population by using the 1960 age-cause specific mortality statistics for white males in the United States. Forty deaths were observed and 18 were expected.

The two papers by Pinto et al. (1977, 1978) refer to the same study and the following is based on the 1978 paper. The cohort studied consisted of 527 men who were living pensioners on January 1, 1949 or who became pensioners before January 1, 1973. Complete job histories were obtained for 525 men. The average duration of employment was 28 years, ranging from 7 to 54 years and beginning in 1910. Death certificates were obtained for all 324 men who had died during the observation period (1949-1973). Expected numbers of deaths were calculated from statistics of the state of Washington.

An exposure index was constructed by using data on urinary levels of arsenic obtained in 1973. Mean urinary concentrations were calculated for 32 departments, and the individual exposure index was obtained by multiplying urinary arsenic level with years of work in a department. If an individual had worked in more than one department, the index values were added. By dividing the exposure index by the total number of years in the smelter, an index of intensity of exposure was obtained, i.e., the average urinary arsenic level. These indices were created to enable interdepartmental comparisons and did not reflect past exposure since air analysis in the 1930s to 1940s indicated that exposure might have been 5-10 times higher at that time. It may have been still higher in 1910.

Data were also obtained on smoking habits from all men still alive and from relatives of men who had died since January, 1961. Table 7-4 (Pinto et al., 1978) shows that there was a significant increase ($p < 0.05$) in deaths from all causes, in cancer deaths in general, and, specifically, in deaths from respiratory cancer. Almost all of the excess mortality could be explained by the increase in lung cancers which could not be explained by smoking.

Table 7-5 shows respiratory cancer deaths in relation to exposure index (a value which reflects both the duration and intensity of exposure). An increase in SMR with exposure is seen. Table 7-6 shows that both duration and intensity of exposure contributed to the excess in respiratory cancer. As stated above, the urine values are relative and do not reflect the actual exposure.

It was also shown that the main excess occurred in ages 65-74 years, whereas at higher ages the lung cancer rate was closer to expected rates.

More recently, Enterline and Marsh (1980, 1982) conducted additional studies on workers at this same copper smelter in Tacoma. A cohort of 2802 males who worked a year or more during the period 1940-1964 was identified. Since a one-year work exposure was required for eligibility into the cohort, actual follow-up did not start until 1941 and extended through 1976. In the cohort, the vital status of 51 could not be verified, leaving 2751 persons. In that group, 1061 deaths had occurred. There was a significant increase in total cancer mortality which wholly depended on an increase in deaths from lung cancer. Arsenic exposure was estimated for each man on the basis of a representative average urinary arsenic level for workers in a given department. Using this representative value, an individual value was calculated for each

TABLE 7-4. OBSERVED AND EXPECTED DEATHS AND STANDARDIZED MORTALITY RATIOS FOR SELECTED CAUSES OF DEATH OF 527 MALES OF COHORT UNDER STUDY*

Cause of death	Disease Classification†	Observed	Expected	SMR
All Causes		324	288.7	112.2‡
Cancer	140-205	69	46.5	148.4‡
Digestive	150-159	20	16.4	122.0
Respiratory	160-164	32	10.5	304.8‡
Lymph, etc.	200-203, 205	2	2.1	95.2
Urinary	180,181	3	3.3	90.9
All Other Cancers		12	14.2	84.5
Stroke	330-334	43	38.0	113.2
Heart Disease	400-443	144	132.3	108.8
Coronary Heart Disease	420	120	110.2	108.9
All Other Heart Disease		24	22.1	108.6
Respiratory Disease	480-493, 500-502	11	10.8	101.8
All Other Causes		57	61.8	92.2

*Cohort consisted of living male pensioners from a copper-smelting plant who were living January 1, 1949, and whose causes of death were noted through December 31, 1973.

†Numbers from rubrics of 7th Revision of International Classification of Diseases.

‡P<.05

Source: Pinto et al. (1978).

TABLE 7-5. OBSERVED AND EXPECTED RESPIRATORY CANCER DEATHS AND STANDARDIZED MORTALITY RATIOS BY ARSENIC EXPOSURE INDEX

			Respiratory Cancer Deaths		
Exposure index	Mean index	No. of men	Observed	Expected	SMR
Under 2,000	1,514	36	1	0.9	111.1
2,000-2,999	2,513	109	4	2.1	190.5
3,000-5,999	4,317	205	11	3.9	282.0*
6,000-8,999	7,473	109	7	2.3	304.3*
9,000-11,999	10,135	38	4	0.7	571.4*
12,000 and over	14,712	29	5	0.6	833.3*

*p <.05

Source: Pinto et al. (1978).

TABLE 7-6. OBSERVED AND EXPECTED RESPIRATORY CANCER DEATHS AND STANDARDIZED MORTALITY RATIOS BY INTENSITY AND DURATION OF EXPOSURE TO ARSENIC

Intensity of exposure	Duration of exposure					
	less than 25 years			25 years and more		
(μg/liter urine)	Observed	Expected	SMR	Observed	Expected	SMR
50-199	2	2.1	95.2	10	3.6	277.8*
200-349	4	1.5	266.7	8	2.2	363.6*
350 and over	3	0.5	600.0*	5	0.6	833.3*

*P<.05

Source: Pinto et al. (1978).

man for each year of employment in a given department, and a total exposure estimate per individual was made by summing values across all jobs and all years of employment.

This method of estimating exposure differed from earlier methods employed by Enterline and co-workers (Pinto et al., 1977, 1978) in that estimates of historic exposure, based upon simple linear interpolations and extrapolations of actual data from 1948-52 and 1973-75, were used to characterize exposure by department, rather than by 1973 urinary measurements. Use of this new method of analysis partially helped to eliminate exposure underestimates of workers employed in the early years of the smelter operation. However, the present method did not totally eliminate this bias because urinary arsenic levels were only determined for workers starting in 1948. For workers exposed prior to 1948 (approximately 80 percent of the present study cohort), urinary arsenic values for 1948-52 were assumed to apply. Furthermore, the average urinary arsenic level for some departments may have been based on only a few samples, thereby limiting the usefulness of the departmental average as a representative measure of any given worker's urinary arsenic level. The authors did note that in areas of the smelter where arsenic levels were reported to be high, workers tended to be measured more often for urinary arsenic. Thus, averages for those areas were, in fact, more representative.

Using this time-weighted measure of exposure, a life table method for accumulative dose, a 10-year lag period and a standard population of mortality rates in the state of Washington, the authors reported that SMRs for respiratory cancer ranged from 155 in the lowest exposure category to 246 in the highest category. Table 7-7 shows the relationship between the time-weighted estimates of arsenic exposure lagged 0 and 10 years and respiratory cancer. This relationship was much weaker than that previously reported by Pinto et al. (1977, 1978). Enterline and Marsh noted that results from the two sets of

TABLE 7-7. RESPIRATORY CANCER DEATHS AND SMRs BY CUMULATIVE ARSENIC EXPOSURE LAGGED 0 AND 10 YEARS, TACOMA SMELTER WORKERS

Cumulative Exposure (μg/As/1) (urine-years)	Lag			
	0 Lag		10-Year Lag	
	Observed Deaths	SMR	Observed Deaths	SMR
<500 (302)	8	202.0	10	155.4
500-1500 (866)	18	158.4	22	176.6*
1500-3000 (2173)	21	203.2**	26	226.4**
3000-5000 (4543)	26	184.1**	22	177.6*
7000+ (13457)	31	243.4**	24	246.2**

* $p<0.05$

**$p<0.01$

()Mean of class interval

Source: Enterline and Marsh (1982).

studies were not totally comparable due not only to the differences in the exposure estimates noted above, but also to differences in follow-up periods. In earlier reports, the follow-up started after exposure stopped at retirement, whereas in the present study, follow-up started at various points in the work experience of the workers, allowing for follow-up and dose accumulation to proceed concurrently. When the authors analyzed a subsample of the present study cohort, consisting of 582 workers retired at age 65 and over (paralleling the experimental design of the earlier studies), a stronger dose-response relationship was, in fact, observed.

Table 7-8 shows the respiratory cancer deaths and SMRs by duration of exposure and time since first exposure. From Table 7-8, it appears that neither duration of exposure nor long latent periods made strong contributions to excess respiratory cancer. The authors suggested that this may have been due to the high SMRs observed shortly after termination of employment but not noted thereafter; thus, for workers with less than 10 years of exposure the

TABLE 7-8. RESPIRATORY CANCER DEATHS AND SMRs BY DURATION OF EXPOSURE AND LATENCY, TACOMA SMELTER WORKERS

	Latency (Years)									
	<10		10-19		20-29		30+		Total	
Duration (years)	Observed Deaths	SMR	Observed Deaths	SMR	Observed Deaths	SMR	Observed Deaths	SMR	Observed Deaths	SMR
<10	1	55.6	10	265.4*	17	210.1**	12	137.9	40	178.9**
10-19	-	-	6	156.4	8	278.0*	4	137.9	18	187.2*
20-29	-	-	-	-	13	197.0*	13	265.3**	26**	226.1*
30+	-	-	-	-	-	-	20	221.3**	20	221.3**
Total	1	55.6	16	210.4*	38	216.3**	49	191.8**	104	198.1**

* p <0.05

**p <0.01

Source: Enterline and Marsh (1982).

SMR is highest 10-19 years after date of hire; for workers employed 10-19 years, the SMR is highest 20-29 years after date of hire, etc. This high relative risk of lung cancer mortality in the decade following termination of employment, which essentially disappears in the following 20 years, prompted the authors to suggest that arsenic may be a promoter rather than an initiator. However, when the authors reanalyzed the data according to a method which followed workers only from the point of termination or retirement, both duration of exposure and intensity of exposure contributed more strongly to respiratory cancer mortality (Table 7-9). The difference in the two analyses led the authors to suggest that the weak dose-response relationship observed in the first analysis may have resulted from a tendency for workers in high-exposure jobs to leave employment more quickly than those in low-exposure jobs.

In studying the interactive effects of sulfur dioxide, the authors did not find significant differences in respiratory cancer incidence in two departments which both had high arsenic exposures (7500 $\mu g/m^3$) but differing SO_2 exposures, one having low to moderate exposures (520 ppm) and the other having essentially none. Because the respiratory cancer SMRs were quite similar in the two departments, the authors suggested that SO_2 exposure did not play an important role in respiratory cancer excess at that particular smelter.

In discussing their overall study results, Enterline and Marsh noted that, in this particular case, a dose response was not observed when dose was measured in terms of cumulative dose. The results seen in Table 7-8 suggest that short exposures seemed to have a disproportionately greater effect than long exposures and that effects of early exposure tended to diminish with time. Table 7-9 suggests that short high-intensity exposures may have a greater effect than longer-term, more low-level exposures. The fact that different results are obtained when different exposure/follow-up methods are

TABLE 7-9. RESPIRATORY CANCER DEATHS AND SMRs BY DURATION AND INTENSITY OF EXPOSURES, TACOMA SMELTER WORKERS

Duration of exposure (years)	Intensity estimate							
	Low†				High++			
	No. at risk	Observed deaths	Expected§ deaths	SMR	No. at risk	Observed deaths	Expected§ deaths	SMR
<10	687	15	8.83	169.9	824	25	12.26	203.9**
10-19	149	7	2.61	268.2*	168	11	3.42	321.6**
20-29	225	10	3.59	278.6**	171	16	2.77	577.6**
30+	159	9	2.98	302.0**	165	11	3.17	347.0**
Total	1220	41	18.01	227.7**	1328	63	21.62	291.4**

* $p < 0.05$.

** $p < 0.01$.

† low = <290 μg/As/ℓ urine (mean 163).

++ high = ≥290 μg/As/ℓ urine (mean 477).

§ Based on Washington State white males.

Source: Enterline and Marsh (1982).

employed suggests that choice of experimental design has a possible influence on results. The possibility that some workers may have simply been more susceptible than others may also have accounted for the dose-response relationships observed in this study.

Because the authors suggested that the role of arsenic in the carcinogenesis process may have been as a promoter rather than an initiator, the significance of cumulative exposure to arsenic as a measure of dose would be questionable in regard to the induction of respiratory cancer as observed in this study. The role of arsenic as an initiator as well as a promoter cannot be ruled out, however. Until further research is done, the authors' conclusions from this study remain speculative.

Mortality of workers in a smelter in Magna, Utah, was studied by Rencher et al. (1977). Average hourly air concentrations of arsenic in 12 work areas were between zero and 22 mg/m^3 in 1975 (NIOSH, 1975). However, exposure before 1959 had likely been higher, since a processing of ore with a relatively low arsenic content began in that year. In the period 1959 to 1969, 965 deaths had occurred among all current or former employees, and death certificates were obtained for virtually all deceased. The proportionate mortality for smelter workers was compared with that for mine workers, concentrator workers, refinery workers, and office workers, and for the population above 20 years of age for the entire state of Utah in 1968. Among the smelter workers, 7 percent of the deaths were due to respiratory cancer, whereas the percentage for the other factory employees varied from 0 to 2.2 percent and was 2.7 percent for the state. Data on smoking were obtained for all smelter workers and for subsamples of the other employee groups and indicated that nonsmoking smelter workers had the same percentage of deaths from lung cancer as mine and concentrator workers who smoked. By applying lifetable methods, age-adjusted death rates were obtained. For smelter workers, a death rate for lung cancer of 10.1 per 10,000 was obtained compared with 2.1 and 3.3 per 10,000 for mine workers and the state population, respectively. Causes of death were compared with a cumulative exposure index obtained by multiplying the number of days spent in each department by the average exposure level and then summing. The average exposure to arsenic as well as sulfur dioxide, sulfates, lead, and copper was found to be higher for the lung cancer cases than for other causes of death.

In addition to these studies, a large study by Lee and Fraumeni (1969) involving 8,047 white males was conducted at the Anaconda Copper Smelter in Montana from 1938 to 1956.* Data were obtained on time, place, and duration of employment for each individual, all subjects having worked at least 12 months in the smelter during the indicated study period. Follow-up was from 1938 to 1964. Death certificates were obtained, and the lifetable method was used to compute the expected number of deaths, using mortality rates of the state of Montana. The smelter workers were classified into 5 cohorts, based on total years of smelter work and the time period in which the years were worked: (1) 15 or more years worked before 1938; (2) 15 or more years worked between 1938 and 1963; (3) 10-14 years; (4) 5-9 years; and (5) 1-4 years. An attempt was also made to classify the workers according to exposure to arsenic and sulphur dioxide. Exposure to arsenic for more than 12 months had occurred among 5,185 men. These workers were divided into three groups; heavy, medium, and light exposures. This division was based on exposure times and place at work. Arsenic concentrations in air were primarily determined from a

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*While the original Lee and Fraumeni study reported on the mortality experience of male smelter workers in unidentified states, subsequent analyses of these workers indicate that they were employed solely at the Anaconda smelter in Montana.

1965 survey by Public Health Service officials (NIOSH, 1975). These air data are shown in Table 7-10.

According to a former public health official associated with this survey, actual measurements were collected during two different periods--one in 1965 and one "about five to six years earlier"--and at several locations within given departments (Archer, 1983; personal communication). It is impossible, however, to determine how the values listed in Table 7-10 are distributed over these different time periods and locations. Further, it is also impossible to determine the number of hours sampled at a given location. Therefore, the arithmetic means used by Lee and Fraumeni to characterize heavy, medium and light exposures can only be viewed as very rough estimates of arsenic exposure, their primary value being in their relative scale of measurement.

In Table 7-11 it is seen that heavy and medium exposures resulted in significant increases in SMR for respiratory cancer. The SMR was highest (800) in workers belonging to cohort 1 (more than 15 years of smelter work before 1938), and then decreased to 263 among workers with 1-14 years of smelter work. Among the workers with light exposure, the SMR was 250, 310, and 214 in cohorts 1, 2, and 3-5, respectively. Among the smelter workers with less than 12 months' exposure to arsenic, the SMR was 286.

Lee and Fraumeni also looked at factors that possibly had confounding effects on their study results. Smoking histories were not recorded; however, based upon information obtained from other studies, Lee and Fraumeni concluded that smoking alone could not have accounted for the excess respiratory cancer mortality of the magnitude observed in their study.

In contrast to smoking, the authors did note a positive relationship between exposure to sulfur dioxide and respiratory cancer mortality; however, they found it difficult to separate the relationship of arsenic from sulfur

TABLE 7-10. 1965 SMELTER SURVEY ATMOSPHERIC ARSENIC CONCENTRATIONS (mg As/m^3)

"Heavy exposure area" as classified by Lee and Fraumeni			
Arsenic Roaster Area			Mean: 1.47
	0.10	0.20	Median: 0.185
	0.10	0.22	
	0.10	0.25	
	0.10	0.35	
	0.10	1.18	
	0.10	5.00	
	0.17	12.66	
"Medium exposure area" as classified by Lee and Fraumeni			
Reverberatory Area			Mean: 1.56
	0.03	0.93	Median: 0.88
	0.22	1.00	
	0.23	1.27	
	0.36	1.60	
	0.56	1.66	
	0.63	1.84	
	0.66	1.94	
	0.76	2.06	
	0.78	2.76	
	0.78	3.40	
	0.80	4.14	
	0.83	8.20	
Treater Building and Arsenic Loading			Mean: 1.50
	0.10	0.48	Median: 0.295
	0.10	0.62	
	0.10	3.26	
	0.11	7.20	
"Light exposure areas" as classified by Lee and Fraumeni			
Copper Concentrate Transfer System			Mean: 0.70
	0.25		Median: 0.65
	0.65		
	1.20		
Samples from Flue Station			Mean: 0.17
	0.10		Median: 0.17
	0.24		
Reactor Building			Mean: 0.004
	0.001	0.003	Median: 0.002
	0.002	0.009	
	0.002	0.010	
	0.002		

Source: National Institute of Occupational Safety and Health (1975).

TABLE 7-11. OBSERVED AND EXPECTED DEATHS FROM RESPIRATORY CANCER, WITH STANDARDIZED MORTALITY RATIOS (SMR), BY COHORT AND DEGREE OF ARSENIC EXPOSURE, 1938-63

Cohort	Respiratory cancer mortality	Maximum exposure to arsenic (12 or more months)* Heavy	Medium	Light
All cohorts combined	Observed	18	44	45
	Expected	2.7	9.2	18.8
	SMR	667†	478†	239†
1	Observed	8	22	14
	Expected	1.0	3.3	5.6
	SMR	800†	667†	250†
2	Observed	6	12	9
	Expected	0.9	2.2	2.9
	SMR	667†	545.†	310†
3-5‡	Observed	4	10	22
	Expected	0.9	3.8	10.3
	SMR	444§	263§	214†
Number of persons in arsenic category*		402	1,526	3,257

*The remaining 2,862 men in the study worked less than 12 months in their category of maximum arsenic exposure and had an SMR of 286†.

†Significant at the 1% level.

‡Cohorts 3, 4, and 5 were combined, since observed and expected deaths were small for each cohort alone.

§Significant at the 5% level.

Source: Lee and Fraumeni (1969).

dioxide. Most of the work areas having heavy arsenic exposure were also areas having medium sulfur dioxide exposure; conversely, all work areas with heavy sulfur dioxide exposure were areas of medium arsenic exposure. The authors did note, however, that persons with the heaviest exposure to arsenic and moderate or heavy exposure to sulfur dioxide were those most likely to die of respiratory cancer in this particular instance.

Since the Lee and Fraumeni study, additional research has been conducted on the employees of the Anaconda smelter. Lubin et al. (1981) studied 5403 of these employees. These workers had all been employed for 12 months or more between January 1, 1938, and December 31, 1956, and were known to be alive as of December 31, 1963. Essentially, this cohort was equivalent to the surviving members from the Lee and Fraumeni cohort.

Exposure was from date hired to December 31, 1963; follow-up was from 1964 to 1977. Classification of exposure categories was similar to that of Lee and Fraumeni. However, unlike the study of Lee and Fraumeni, a cumulative arsenic exposure index was calculated for each worker. This index was derived by, first, weighting the three exposure categories and then multiplying the number of years an individual worked in a given category by this weight and summing over categories. The weights were derived from mean airborne dust concentrations taken during 1943 to 1958, which averaged 11.3, 0.58 and 0.29 mg As/m^3 in the respective heavy, medium and light categories. These values differed from those used by Lee and Fraumeni to group departments (Table 7-9). It should be noted that the exposure estimates used by Lee and Fraumeni were based upon more recent monitoring data primarily collected in 1965. Lubin et al. reduced weights in the heavy category by a factor of 10 in order to account for the wearing of respirators as was observed "at least in recent years." SMRs were calculated by comparisons to U.S. white males.

The mortality experience of workers during the years 1964 to 1977 was similar to that of the workers studied by Lee and Fraumeni during the period 1938-1963. Excess deaths from respiratory cancer corresponded to areas of highest arsenic levels. The authors noted an overall strong gradient in risk associated with the indices of cumulative arsenic exposure. However, the authors also noted that this gradient was less clear when weighting of the high-exposure category was reduced ten-fold to account for respirator usage. According to Welch et al. (1982), however, respirator usage was not common prior to 1964.

Some study differences were noted by Lubin et al. between their respective study and that of Lee and Fraumeni. Differences in excess respiratory cancer--65 percent in the more recent period versus a three-fold excess in the earlier period--were partially attributed to differences in respiratory cancer rates observed between the two comparison populations. Respiratory cancer rates in the general populace have increased in recent years; therefore, comparisons to recent populations will produce lower relative risks than comparisons to past general populations in which the rates of respiratory cancer were lower. The authors also noted that the comparison populations differed in composition. Lee and Fraumeni used white males in the state of Montana as their standard population, whereas Lubin et al. used U.S. white males. As noted in this study, as well as elsewhere (Welch et al., 1982; Higgins et al., 1982), death rates for specific causes of death (inclusive of respiratory cancer) have been reported to be lower in Montana. Finally, the authors suggested the possibility that individuals most susceptible to lung cancer contracted the disease in the earlier period and were, thus, lost to the study follow-up due to death.

In looking at SO_2 exposures, the authors were unable to totally separate the possible interactive effects of SO_2 with arsenic. However, they did note that after controlling for arsenic, no significant increase in mortality could be associated with heavy or medium SO_2 exposures, whereas the association with arsenic exposure persisted after controlling for SO_2. This finding is consistent with that of other researchers studying smelter populations (Enterline and Marsh, 1982; Welch et al., 1982; Higgins et al., 1982).

In an update of the earlier study co-authored with Fraumeni, Lee-Feldstein (1983) observed the mortality experience of the same Anaconda workers (with the exception of two women) from 1938 to 1977. The workers (8045) were assigned to one of five cohorts on the basis of total years of employment. Cohort 1 worked 25+ years; cohort 2, 15-24 years; cohort 3, 10-14 years; cohort 4, 5-9 years, and cohort 5, 1-4 years. SMRs were calculated by comparison to the combined white male populations in the states of Idaho, Wyoming and Montana.

Of the thirteen specific causes of death considered, tuberculosis, digestive and respiratory cancer, vascular lesions of the CNS, diseases of the heart, emphysema, and cirrhosis of the liver showed a significant excess of observed deaths over that expected; however, only excesses in respiratory cancer showed a positive gradient with length of employment when comparing cohorts 1 through 5. The ratio of observed to expected mortality from respiratory cancer was approximately 5.1, 4.5 and 2.3 in the heavy, medium and light arsenic-exposure groups, respectively. This was in accord with the earlier results of Lee and Fraumeni, except that the ratio of observed respiratory cancer deaths to that expected in the heavy exposure category in the earlier study was 7.

Brown and Chu (1983c) discussed the Lee-Feldstein (1983) data with regard to its implications on the multistage theory of cancer. They indicated that analysis of the data suggests that arsenic acts as a late-stage carcinogen because: (1) there was an increasing excess lung cancer mortality risk with increasing age at initial exposure, and (2) the excess mortality was independent of time after exposure stopped.

Higgins et al. (1982) and Welch et al. (1982) reported on a sample of 1800 workers at the Anaconda smelter. Compared to the 8047 workers studied by Lee and Fraumeni, this cohort of 1800 workers included all the workers originally designated in Lee and Fraumeni's heavy exposure category (277) and a random sample (20 percent) of the remaining known workers. The date of entry into the study cohort ranged from 1938 to 1956, providing the individual had one year of work experience. Unlike other studies on these workers, smoking histories on the 1800 workers were obtained either by direct questioning or by proxy respondent. SMRs were based on comparisons to white males both in the state of Montana and the United States.

From industrial hygiene records for the period 1943 to 1965, estimates of airborne arsenic concentrations within 35 smelter departments were provided. A total of 818 samples were collected from 18 departments and departmental averages were calculated from these measurements. The remaining 17 departments were estimated by analogy with those that were known. The departments were then grouped into four categories in which arsenic exposure was characterized as low (<100 $\mu g/m^3$), medium (100-499 $\mu g/m^3$), high (500-4999 $\mu g/m^3$) or very high (>5000 $\mu g/m^3$).

Workers were assigned to these categories based upon estimations of both time-weighted average (TWA) arsenic exposure levels and ceiling levels. TWA values were individually calculated based upon the time that a worker spent in a given department and the average arsenic concentration estimated for that department. This quantity was summed across all departments the individual worked in and was divided by total time worked to yield a TWA. TWA arsenic exposures were calculated at entry into the study cohort and at the beginning of January, 1964, corresponding to the end of the follow-up period used by Lee and Fraumeni. TWA differences between these two periods could have increased, decreased, or remained the same depending on the work history of an individual worker. In contrast, ceiling levels were defined as the highest level to which an employee was exposed for a period of 30 days or more. Ceiling levels were calculated at entry into cohort, at the beginning of 1964 and at the beginning of 1978. Unlike a TWA value, a worker's ceiling level could only increase or remain the same from point of entry into the cohort.

Data were analyzed according to five different exposure/follow-up methods which varied in the amount of overlap allowed between exposure and follow-up periods. Method I, the primary method used by the authors, included each worker's arsenic exposure up to the date he entered the cohort. Follow-up was from entry to 1978; thus, there was no overlap of the two periods. Method IV--exposure from date hired to 1964, follow-up from 1964 to 1978--also had no overlap. Methods II and V had complete overlap--exposure from date hired to 1964, follow-up from 1938 to 1964 and exposure from date hired till termination, follow-up from 1938 to 1978, respectively. Method III had partial overlap--exposure from date hired to 1964, follow-up from 1938 to 1978. Except where specifically noted, results were given according to Method I.

The results of the study supported the thesis that exposure to arsenic was strongly related to respiratory cancer mortality in workers at the Anaconda smelter. SMRs for the total cohort for all causes of death were identical when compared to either the state of Montana or U.S. white males (both SMRs = 133, significant at 0.01 level); however, SMRs for specific causes were somewhat higher when compared to Montana males than when compared to U.S. males. Exposure to arsenic appeared to be the principal factor in the observed increased risk for respiratory cancer, the study cohort having 3 times the expected death rate for white men living in Montana. Exposure to other occupational contaminants, such as sulfur dioxide and asbestos, did not appear to account for respiratory cancer excess, while smoking explained only a small fraction of the excess.

Of particular note were the differing respiratory cancer results obtained under the two categories of arsenic exposure (Tables 7-12 and 7-13). The SMR for men in the lowest TWA category was elevated, although not significantly so, whereas the SMRs in the other TWA categories were significantly elevated. Mortality for respiratory cancer by ceiling arsenic exposure showed that SMRs were only signficantly elevated in the high and very high categories, whereas they were close to expectation in the two lower exposure categories. Date of hire showed a definite relationship to mortality from respiratory cancer. Workers who were employed in the early years of the smelter operation--from 1884 to 1938, but particularly prior to 1923, when a selective floatation process which markedly improved fume and dust recovery was introduced--had higher SMRs, indicating that the overall higher arsenic exposures in the early years were associated with higher death rates from respiratory cancer. Age at hire did not seem to have a confounding effect, although the authors did note that the SMRs for respiratory cancer showed more fluctuation with age than did all causes of death.

Tables 7-14 and 7-15 show comparisons of TWA and ceiling respiratory cancer mortality as analyzed by the different exposure/follow-up methods. The authors noted that, while there was some variation in the SMRs derived from the different methods, the same basic pattern of increasing respiratory cancer SMRs with increasing TWA and ceiling arsenic exposures could be seen for each method. The authors indicated that none of the exposure/follow-up methods found a significantly ($P < 0.05$) increased respiratory cancer risk for ceiling level

TABLE 7-12. MORTALITY FOR ALL CAUSES AND RESPIRATORY CANCER FROM 1938 TO 1978 BY TIME-WEIGHTED AVERAGE (TWA) ARSENIC EXPOSURE AS OF ENTRANCE INTO COHORT

TWA Arsenic ($\mu g/m^3$)	N	Person-Years	All Causes			Respiratory Cancer		
			Obs	Exp	SMR	Obs	Exp	SMR
<100	547	13152	219	196.7	111*	11	7.9	138
100-	542	14157	216	178.0	121**	22	7.3	303**
500-	565	13460	292	184.7	158**	29	7.7	375**
5000-	146	3552	89	56.1	159**	18	2.6	704**

* Significant at 0.05 level

** Significant at 0.01 level

Source: Higgins et al. (1982).

TABLE 7-13. MORTALITY FOR ALL CAUSES AND RESPIRATORY CANCER BY CEILING ARSENIC EXPOSURE AS OF ENTRANCE INTO COHORT

Ceiling Arsenic ($\mu g/m^3$)	N	Person-Years	All Causes			Respiratory Cancer		
			Obs	Exp	SMR	Obs	Exp	SMR
<100	445	10591	165	152.1	108	8	6.2	129
100-	276	7083	80	80.5	99	4	3.4	116
500-	833	20757	416	288.5	144**	41	11.8	348**
>5000	246	5889	155	94.4	164**	27	4.1	662**

* Significant at 0.05 level

** Significant at 0.01 level

Source: Higgins et al. (1982).

TABLE 7-14. RESPIRATORY CANCER MORTALITY BY METHOD OF ANALYSIS AND TWA ARSENIC CATEGORY

TWA Arsenic (μg/m³)	METHOD OF ANALYSIS I[a]				III[b]				IV[c]			
	N	Obs	Exp	SMR	N	Obs	Exp	SMR	N	Obs	Exp	SMR
<100	547	11	7.9	138	522	5	7.9	63	354	4	4.8	84
100-	542	22	7.3	303**	580	28	7.8	359**	410	16	5.1	313**
500-	565	29	7.7	375**	515	29	7.1	408**	313	9	4.2	216*
≥5000	146	18	2.6	704**	183	18	2.7	673**	111	9	1.6	573**

*Significant at 0.05 level

**Significant at 0.01 level

[a] Exposure from date hired to cohort entry. Follow-up from cohort entry to 1978.

[b] Exposure from date hired to 1964. Follow-up from cohort entry to 1978.

[c] Exposure from date hired to 1964. Follow-up from 1964 to 1978.

Source: Higgins et al. (1982).

TABLE 7-15. RESPIRATORY CANCER MORTALITY BY METHOD OF ANALYSIS AND CEILING ARSENIC CATEGORY

Ceiling Arsenic ($\mu g/m^3$)	METHOD OF ANALYSIS I[a]				III[b]				IV[c]				V[d]			
	N	Obs	Exp	SMR	N	Obs	Exp	SMR	N	Obs	Exp	SMR	N	Obs	Exp	SMR
<100	445	8	6.2	129	275	3	4.0	75	178	3	2.3	130	267	3	3.9	77
100-	276	4	3.4	116	218	2	2.5	79	152	0	1.6	-	210	2	2.4	83
500-	833	41	11.8	348**	970	41	13.8	298**	656	20	8.7	230**	969	41	13.7	300**
≥5000	246	27	4.1	662**	337	34	5.2	652**	202	15	3.0	496**	354	34	5.5	617**

*Significant at 0.05 level

**Significant at 0.01 level

[a] Exposure from date hired to cohort entry. Follow-up from cohort entry to 1978.

[b] Exposure from date hired to 1964. Follow-up from cohort entry to 1978.

[c] Exposure from date hired to 1964. Follow-up from 1964 to 1978.

[d] Exposure from date hired to 1978. Follow-up from cohort entry to 1978.

Source: Higgins et al. (1982).

categories less than 500 μg/m^3 of arsenic (Table 7-15). For the respiratory cancer SMR analysis by TWA exposure, a significantly ($P < 0.05$) elevated SMR was not found in the lowest dose category. In the 100-499 μg/m^3 TWA exposure category, there was a significantly ($P < 0.05$) elevated respiratory cancer SMR (Table 7-14). The respiratory cancer SMR in this category would not have been significant, however, if those respiratory cancer deaths with <500 μg/m^3 ceiling arsenic exposure had been excluded from the analysis. The authors concluded that if a worker in the study had not been exposed to <500 μg/m^3 ceiling level, he would have had little, if any, excess cancer risk.

A number of criticisms have been made of this study (48 FR 1864), a few of which bear mentioning at this point. The representativeness of the exposure estimates as they relate to overall exposure conditions in the smelter and specifically to the cohort's TWA and ceiling exposures has been criticized. The authors noted that individual worker exposure estimates, based upon area measurements rather than personal samples, would not likely have great precision. Furthermore, estimates of analogy--as in 17 of the departments--weaken the reliability of overall exposure estimates, although areas that were thought to be "problem" areas of high exposure were areas that were generally measured. The extent to which respirator usage was an effective protective measure is also unknown. In interviews with former workers, however, indications were that the use of respirators was not widespread or regular during the period prior to 1964. Collectively, the authors stated that exposure estimates probably tended to affect results such that exposure values were underestimated from 1884 to 1938, were reasonable for 1938 to 1964, and were overestimated for the period after 1964.

The authors found that workers who had not experienced a ceiling level of 500 μg/m^3 of arsenic did not have an elevated risk of lung cancer. However, the number of lung cancer deaths that could be predicted using a linear dose-response model (see Section 7.3.1) in the group of workers with <500 μg/m^3 arsenic ceiling exposure would not be significantly different than the number of lung cancer deaths actually found. This was determined using the linear risk model (Section 7.3.1) and data obtained from Higgins and made available by Consultants in Epidemiology and Occupational Health (1982) on the cross-tabulation of the number of workers in cumulative and ceiling exposure categories. Higgins et al. are presently undertaking a study of the entire Anaconda cohort; however, results of this study will not be forthcoming until late 1984.

In foreign smelters, an excess in lung cancer mortality has also been found. The Rönnskär smelter in Sweden, which has been processing arsenic-rich ores since the 1920s, has been the subject of several studies. Axelson et al. (1978) conducted a case-control study of mortality from respiratory cancer in relation to employment at that smelter. In the parish surrounding the smelter, 369 deaths have been recorded in the registers for men aged 30-74 years during the years 1960 to 1976. Causes of death were obtained in all cases. Smoking habits were obtained from medical files. Cases were defined as subjects who died of malignant tumors of the lung, other cancers, cardiovascular disease, cerebrovascular disease, and cirrhosis of the liver. The control group was made up of persons from the same parish who died from causes other than the above, excluding 44 persons with diabetes, mental deficiency and unclear diagnoses. Attempts were made to assess exposure and four exposure groups were constructed, based on intensity and duration of exposure and time between initiation of exposure and death. It was found that occupational exposure to arsenic was associated with a significant increase in deaths from lung cancer. For the three exposure groups with at least 3 months of exposure occurring 5 years prior to death, the lung cancer mortality ratio was 4.6. In these groups, there was an increase with exposure intensity. For the fourth group in which either persons were not exposed or the first exposure was for a period less than 3 months and/or death occurred within 5 years of this first exposure, a risk of respiratory cancer was not found.

Other agents, including sulfur dioxide, did not seem to be associated with lung cancer in this study. In 83 percent of the exposed lung cancer cases there was a history of smoking. A study of smoking habits by Pershagen (1978) showed that the excess lung cancer mortality could not be explained by smoking habits.

Because a high lung cancer mortality rate was noted among males in Saganoseki-machi, Japan, for the period 1967-69, Kuratsune et al. (1974) did a case-control study of lung cancer cases in that town. The nineteen cases of lung cancer for the period 1967-69 were compared with nineteen controls randomly selected from deaths of diseases other than cancer of the lung, skin, or bladder for the 1967-69 period. Smoking and drinking habits, residential and occupational histories, and exposure to atomic bomb radiation were the factors compared. Fifty-eight percent of lung cancer cases were found to be former smelter workers vs. 15.8 percent in the controls ($p < 0.01$). The relative risk was reported to be 9.0 (confidence limits not reported). No difference was found between the cases and controls for smoking habits, residential history, drinking habits, or atomic bomb radiation.

Tokudome and Kuratsune (1976) did a cohort study of 2765 male workers, including 839 copper smelter workers at the metal refinery in Saganoseki-machi, Japan. Deaths which occurred between 1949 and 1971 were analyzed in the study. The expected number of deaths was calculated using mortality data for Japanese males. A significantly increased mortality was noted for lung cancer (SMR = 1189; observed = 29; expected = 2.44; $p < 0.01$) and colon cancer (SMR = 508; observed = 3; expected = 0.59; $p < 0.05$). A dose response was demonstrated between lung cancer mortality and the degree of exposure measured by length of employment and level of exposure. A very high excess mortality from lung cancer (SMR = 2500; 10 observed; 0.4 expected; $p < 0.01$) was found among smelter workers who had worked in the heaviest exposure category and who had been employed over 15 years before 1949. The latent period in this study ranged from 13 to 50 years.

Studies have also been performed to see if there is any increase in lung cancer among residents in areas surrounding smelters. Lyon et al. (1977) compared the incidence of lung cancer and lymphoma in Utah residents from 1970-1975 in relation to the distance of these residents from a smelter and found no association. It should be noted that distance from the smelter was based on address at the time of diagnosis; nothing regarding the length of time lived near the smelter was factored into the analysis. Furthermore, use of lymphoma cases as controls is questionable since lymphomas may also be associated with arsenic (Ott et al., 1974). Thus, the conclusion of the study that there was no association between lung cancer and distance from the smelter is questionable.

In a more recent study, Rom et al. (1982) compared lung cancer with breast and prostate cancer in residents living near a non-ferrous smelter in El Paso, Texas. The study period ranged from 1944-1973. Similar to methods used by Lyon et al. (1977), comparisons were made in relation to distance from the smelter. Breast and prostate cancer were chosen as control cancers because they have no known association with arsenic exposure. The authors reported that the distribution of lung cancer cases (575) and control cancer cases (1490) was roughly the same for the different distances studied. No association between lung cancer and distance from the smelter was found, nor were there any associations for race, age or sex. However, the authors did note that they were unable to control for such factors as smoking, occupation and migration and were unable to obtain environmental exposure measurements over most of the years studied.

In Montana, the studies by Newman et al. (1976) showed that there was an increase in the incidence of lung cancer among men in cities where copper smelters are located. In one of the cities there was also an increased incidence of lung cancer among women. It was not stated to what extent occupational exposure had caused the increase, and there was no control for smoking.

In the Blot and Fraumeni study (1975), the cancer mortality in 71 counties with smelters and refineries was studied from 1950 to 1969. Comparisons were made with the remaining 2,985 counties in 48 states. In 36 counties with smelters processing copper, lead, or zinc ores, lung cancer mortality was significantly higher both among males ($p < 0.01$) and females ($p < 0.05$). For all 36 counties with these industries the median SMR was 112 for males and 110 for females. Although occupational cancers were included in the analyses and, therefore, contributed to some of the excess risk, the number of workers in the smelter industry generally comprised a small fraction of the total population (less than 1 to 3 percent); thus, it was unlikely that occupational exposures alone accounted for the observed excess mortality in males or the increased risk for females. Although this study is suggestive of a lung cancer effect in populations surrounding smelters, it is unknown if the lung cancer cases were even exposed to arsenic. Thus, the results, although suggestive, are inconclusive.

In the Baltimore, Maryland area, Matanoski et al. (1976, 1981) studied cancer mortality in areas near the earlier mentioned pesticide facility. They found an excess of all cancers and respiratory cancers among males in the area nearest to the factory but not among females. Soil arsenic levels generally corresponded to lung cancer and all cancer incidences, with the highest average of arsenic in the soil (63 ppm) reported in the area with the greatest cancer risk. The authors were unable to account for the differences noted between males and females, but suggested that smoking may have contributed to these differences. Other environmental factors and/or occupational exposure may have also influenced cancer mortality excess. Further environmental sampling of arsenic and conducting of community surveys were recommended to address these unknown factors.

Pershagen et al. (1977) studied cancer deaths in the area surrounding the Rönnskär smelter in Sweden. From 1961 to 1975 there was a significant excess of respiratory cancers in the male population (SMR = 250) compared to residents in an area without known emissions of arsenic but in which the same age distribution and occupational profiles applied. When the occupationally exposed cases were excluded, significant increases in respiratory cancers were no longer detected (SMR 173); however, the male population still showed a tendency to excess lung cancers. No similar occupational group was excluded from the comparison population, however. There was no tendency to an increase in lung cancer among women.

7.1.2.2 Cancer of the Skin and Precancerous Skin Lesions--An elevation in the proportionate mortality of skin cancer was reported by Hill and Faning (1948) for factory workers manufacturing sodium arsenite and by Roth (1957, 1958) for German vintners. In addition, an increased incidence of skin cancer has been reported after long-term oral exposure to arsenic.

In Taiwan a large population had long-term exposures to inorganic arsenic in drinking water. Exposure started in 1910-20 when water was obtained from deep wells, 100-200 m. below the surface. Already in the 1920s, vascular changes began to appear, and in the 1950s, the first epidemiological studies were conducted. The arsenic content of the water varied from 0.01 to 1.82 mg/l (Ch'i and Blackwell, 1968; Astrup, 1968; Tseng et al., 1968; Tseng, 1977), generally being 0.4 - 0.6 mg/l, whereas water from shallow wells or other surface waters generally contained from near zero to 0.15 mg As/l.

Tseng et al. (1968) and Tseng (1977) have reported the results from a large-scale epidemiologic survey of arsenic-related diseases in an area with high arsenic concentrations in drinking water. The population at risk was 103,514 persons. Thirty-seven villages with a population of 40,421 were surveyed house-to-house. Examinations were made with special attention to pigmentation, hyperkeratosis, and cancer of the skin. Four hundred twenty-eight cases of arsenical skin cancer were found, resulting in a rate of 10.6/1000. No cases were under 20 years of age. The prevalence rate increased markedly with age except for females >70. Over 10 percent of the people >59 were affected by skin cancer. The overall male/female ratio was 2.9:1, with males having a higher rate in all age groups >29.

The villages were divided into 4 exposure levels: <0.3, 0.3-0.6, ≥0.6, and undetermined, based on their water arsenic content. There was a clear cut ascending gradient of prevalence from low to high in each of 3 age groups (Table 7-16).

Hyperpigmentation (melanosis) was found in 18.4 percent of the total population: 19.2 percent for males and 17.6 percent for females. Usually the prevalence was higher for males than females. The rates increased steadily with age for males and did likewise for females until a peak was reached at 50-59, followed by a gradual decline.

TABLE 7-16. PREVALENCE OF SKIN CANCER (per 1000) BY AGE AND ARSENIC EXPOSURE (ppm)

Arsenic content of drinking water (ppm)	Age 20-39	Age 40-59	Age >60
<.3	1.3	4.9	27.1
0.3 - 0.6	2.2	32.6	106.2
>0.6	11.5	72.0	192.0

Source: Adapted from Tseng et al. (1968).

The overall prevalence rate for keratosis was 7.1 percent - 7.5 percent for males and 6.8 percent for females. Males had higher rates in the greater than-49-year group. The prevalence increased for both males and females up to age 70 and then declined.

As was the case for skin cancer, the prevalence rates for hyperpigmentation and keratosis suggested that positive correlations existed between these conditions and the arsenic content of the water in the artesian wells; the greater the arsenic content, the higher the prevalence.

In the total survey of 40,421 people, 7418 cases of hyperpigmentation, 2868 of keratosis, 428 of skin cancer, and 360 of Blackfoot disease (see Section 5.2.2) were found. Many of these occurred in combination in the same individual.

The data were examined by comparing expected (based on overall rates) and observed rates for various combinations of the 4 end points. The obtained ratios indicated quite strongly that a common underlying cause existed for the 4 conditions, presumably chronic arsenicism.

A control population of 7500 persons from nonendemic areas was examined in the same way as the arsenic-exposed persons. Four thousand, nine hundred and seventy-eight people lived on Matsu Island; its water supply was from shallow wells and no arsenic was detectable by the analytic methods used in the main series. The remaining portion of control population members came from 5 villages on Taiwan whose water source was shallow wells with arsenic levels between 0.001-0.017 mg/ℓ (ppm). No cases of melanosis, keratosis, or skin cancer were observed in the entire control population.

Although age at onset of the conditions was difficult to assess, some information on latency was obtained. "We know from the study that the youngest cancer patient was 24, the youngest with hyperpigmentation was 3, and the youngest with keratoses was 4." This meant that hyperpigmentation could occur in patients who were exposed from birth for at least 3 years, keratosis for 4 years, and cancer for 24 years (Tseng et al., 1968).

While the Taiwanese data collected in the late 1960's strongly implicated arsenic as the etiological source of the observed diseases, recent findings have called to question the hypothesis of arsenicism as sole causative source in the induction of cardiovascular effects. The discovery by Lu et al. (1977b, 1978) of fluorescent compounds identified as alkaloids--either lysergic acid, dihydrolysergic acid or a derivative of ergotamine tartfate--has opened the possibility that other toxic mechanisms may have been involved. Ergotamine-like compounds in combination with high alkalinity, characteristic of these waters, have been shown to cause gangrene (Lu et al., 1977a,b; 1978). Whether these compounds have a confounding effect on skin cancer is presently unknown.

In addition, recent analyses by Irgolic (1982) on a limited number of samples have shown the water samples to contain predominantly pentavalent arsenic and no organic arsenicals. Samples taken from two wells in the Yenshei Province of Taiwan were collected in plastic cubitainers. Two samples each of unpreserved water and water preserved either by addition of 0.1 weight percent of ascorbic acid or by acidification of 0.1 M HNO_3 were sent to the U.S. for analysis a few days after collection. Treatment of samples by HNO_3 or ascorbic acid was done immediately upon collection (Irgolic, 1983; personal communication). Samples were analyzed during a two-week period after arrival in the U.S., the total time lapse between collection and analyses ranging from one to three weeks. The results of the analyses can be seen in Table 7-17. Additional water samples collected from other parts of Taiwan also contained pentavalent arsenic. However, these samples were less reliable in that the collection period was unknown, and, upon arrival in the U.S., these samples had a yellowish hue with some flocculated matter present (Irgolic, 1982).

Several questions still remain to be answered in regard to water-usage patterns of the Taiwanese. For instance, it is not clear how quickly the water drawn from the wells was consumed; nor is it clear how much of the water was consumed in tea or other beverages where cooking preparations, such as boiling, would have altered the chemical form of the arsenic. These questions need to be answered in light of the possible effect these answers might have on interpreting the observed skin cancer incidence.

Ch'i and Blackwell (1968) conducted a case-control study in the area of Taiwan studied by Tseng et al. The authors compared a variety of factors between 353 cases of Blackfoot disease and 353 controls matched for sex and age. Socioeconomic status, occupation, cigarette smoking, diet, and consumption of deep well water which was arsenic-contaminated were the factors compared. Two factors were found to be significantly different between the cases and controls. Significantly ($P < 0.01$) more cases than controls were found to consume deep well water and to have a lower socioeconomic status. Though the author concluded that the primary contributing cause of Blackfoot disease was consumption of deep well water, the socioeconomic differences cannot be completely discounted.

TABLE 7-17. RESULTS OF TOTAL ARSENIC ANALYSES AND ARSENITE AND ARSENATE DETERMINATIONS IN THE YENSHEI WATER SAMPLES*

				TOTAL ARSENIC, ppm			
Sample	Ascorbic Acid 0.1%	0.1 M HNO_3	Un-preserved	HG[a]	ICP[b]	Arsenite, ppm by HG	Arsenate, ppm by HG
Well I	-	+	-	0.84±0.02	0.73±0.01	-	-
	+	-	-	0.88±0.01	0.72±0.01	0.024±0.001	0.85±0.01
	-	-	+	0.85±0.01	0.73±0.01	0.022±0.001	0.83±0.08
Well II	-	+	-	1.05±0.08	0.95±0.01	-	-
	+	-	-	1.16±0.03	0.96±0.01	-	-
	-	-	+	1.11±0.02	0.98±0.01	0.024±0.01	1.08±0.02

*Average ± average deviation of at least three determinations per sample.

[a] HG = Hydride generation technique.

[b] ICP = Inductively coupled argon plasma emission spectrometry.

Source: Irgolic (1982).

Other studies have also explored the relationship of arsenic to skin cancer and various skin lesions. Similar chronic effects as seen in Taiwan have been reported in other countries.

In Antofagasta, Chile, a new water supply was put into service in 1958. In the 1960s, physicians began noticing dermatological manifestations and even deaths, especially among children. In an investigation of skin pigmentation in 27,088 school children from the province of Antofagasta, an overall incidence of 12 percent was reported (Borgono and Greiber, 1972). It was discovered that the drinking water contained 0.8 mg As/l. Borgono and Greiber compared 180 inhabitants of Antofagasta with a community without exposure to arsenic via drinking water. Abnormal skin pigmentation was reported to be present in 80 percent and hyperkeratosis in 36 percent of the Antofagasta inhabitants, whereas none was found in the control group ($p \leq 0.05$).

In 1970, a water treatment plant was installed and there was a considerable drop in arsenic. According to Borgono et al. (1977), there were no skin lesions in children born since the water treatment began. However, it should be noted that in this more recent study the sample size of children born since the water treatment plant began was small (306) and no comparison population was studied. Therefore, any conclusions associating the lack of dermatological manifestations with the decrease in arsenic must take into account the small sample size. In regard to skin cancer, the follow-up may not have been long enough to detect a difference.

In addition to the work of Borgono and co-workers, Zaldivar and Guillier published a series of papers on the Antofagasta situation (Zaldivar, 1974; Zaldivar, 1977; Zaldivar and Guillier, 1977). The first of these (Zaldivar, 1974) describes a study on a total of 457 patients (208 males and 249 females) bearing cutaneous lesions (leukoderma, melanoderma, hyperkeratosis, and squamous cell carcinoma). The cases were collected both by the author and the local hospital during the period 1968 to 1971. Children 0 to 15 years of age accounted for 69.2 percent of the male cases and 77.5 percent of the female cases. The average incidence/100,000 for cases with cutaneous lesions in 1968 to 1969 were 145.5 and 168.0 for males and females, respectively. By 1971 the incidence rates had dropped to 9.1 and 10.0 for males and females, respectively. The decline in morbidity was so rapid that a conclusion that arsenic was the cause of the skin lesions would have to be considered suspect.

The existence of arsenical waters in an eastern area of the province of Cordoba, Argentina, has been known for many decades (Arguello et al., 1938; Bergoglio, 1964). Effects noted on the population from this area include hyperpigmentation, keratosis, and skin and respiratory cancer. A large area of the province, mainly in the east and somewhat to the south, is the focal point for chronic endemic regional arsenical intoxication (CERAI) (Arguello et al., 1938). This CERAI is due to the ingestion of well water coming from the uppermost sheet of underground water--the principal source of arsenic--as well as to the ingestion of arsenic from the wells, which varies among wells throughout the region. The concentration also varies with rainfall. Vanadium is also elevated in areas with high arsenic content. A later report (Bergoglio, 1964) indicates that progress had been made in improving the hygienic condition of the drinking water.

Arguello et al. (1938) summarized the early history of investigations (1913 to mid-1938) into the health outcomes associated with the ingestion of arsenic-contaminated water. They also reported the results of an investigation of a large series of epitheliomas collected from mid-1932 to September 1938 at the dermatosyphilology clinic of a medical school in the arsenic-contaminated area. The series consisted of 323 cases of epithelioma of which 39 or 12.1 percent were cases with clinical evidence of CERAI.

Patients exhibiting CERAI had characteristic cutaneous lesions, represented by the symmetrical palmo-plantar keratoderma and melanoderma, although the latter symptom was seen less frequently. Foot and hand keratoderma were seen in 100 percent of the CERAI patients. Appearance usually occurred 2 to 3 years after the onset of intoxication. A prodrome of the keratosis is the appearance of an erythema in the same location, and a subjective crawling sensation and local fever. Otherwise, it is a state of dryness which precedes the keratosis.

Arguello et al. (1938) also reported that most patients also had hyperhidrosis and abnormalities of pigmentation. The melanoderma appeared early in the process and was variable among patients. It was described as small dark spots ranging in diameter from 1 to 10 mm. They had a tendency to coalesce and appeared predominantly on the trunk in the areas not exposed to the sun. Atropy was associated with telangiectasia and loss of color, or leukoderma, between the hyperpigmented areas (the "raindrop" appearance cited by Reynolds, 1901).

Geographically, the largest proportion of cases in the clinic came from the areas with the highest incidence of CERAI. Because the authors' data are not population-based, however, it cannot be stated that the incidence of skin cancer is significantly increased in these areas. The authors reported that the hands and feet were the locations of choice for the arsenical epitheliomas (38.5 percent vs 3.9 percent) compared to the nonarsenical epitheliomas. An example of this was seen in the head where 81.6 percent of the nonarsenical epitheliomas occurred versus 15.4 percent for arsenical epitheliomas.

Bergoglio (1964) did a proportionate mortality study of residents of certain departments (counties) in the Province of Cordoba, Argentina, where endemic arsenic levels in the water supply are reported to be very high. The proportion of cancer deaths was higher in those departments than for the province as a whole (23.84 percent versus 15.3 percent; $P < 0.05$). Of the cancer deaths, respiratory cancer constituted 35 percent and skin cancer 2.3 percent. From the description by the author, it can probably be inferred that these data are not age-adjusted. No comparison is made of the percentage of respiratory and skin cancer deaths in the affected departments with the respective proportion for Cordoba Province.

Morton et al. (1976) investigated the relationship of skin cancer morbidity and the ingestion of arsenic-contaminated drinking water in Lane County, Oregon. The southcentral region of Lane County is underlaid by an arsenic-rich stratum called the Fisher formation, which is known to produce high arsenic levels in waters from wells drilled into the land. An extensive search of the pathology records of medical providers in Lane County, Oregon, was conducted in 1972 for the occurrence of skin cancer during the years 1958 to 1971. Cases were thoroughly screened to eliminate duplications and were then coded to 1970 census tract numbers according to the residential address at the time of diagnosis. Water samples were obtained in all census tracts at selected points in all municipalities and water districts in the county, as well as from single-family water sources. The single-family water sources were neither randomly nor uniformly distributed throughout the county but instead were heavily concentrated in the regions believed to have water arsenic problems. Water samples collected during 1968 to 1974 were compiled into mean concentrations for each census tract and census tract region. The authors stated that it seemed reasonable to assume that the samples were representative of the earlier time periods as well.

The skin cancer data were expressed in four sets of rates because of the availability of 4 sets of population estimates. Overall mean annual incidence rates for the entire 1958 to 1971 period used mean population estimates based on all four sets of denominators. Census tract regions were devised to simplify analysis and presentation. Table 7-18 presents the water arsenic level obtained. As can be readily seen, the South rural area had a much greater arsenic exposure than any of the other sections of the county. Figure 7-1 contrasts the parts of the county underlaid by the Fisher formation, and subsequent higher arsenic levels, with the parts of the county experiencing higher squamous cell skin cancer. Relatively little concordance is noted. A multiple regression analysis performed by the authors demonstrated essentially no relation between skin cancer and water arsenic. It should be noted that water arsenic levels in this study varied from 0 to 2150 ppb. The authors point out that the Lane County water arsenic levels were much lower than those reported for Taiwan and Antofagasta. In particular, only 5 percent of the Lane County samples contained 100 or more ppb of arsenic in contrast to 48 percent of the samples in that range in the Taiwan data.

Similar findings were reported by Southwick et al. (1981) in a study conducted on residents of West Millard County, Utah. As an area in which naturally occurring arsenic in public drinking water had been reported, West Millard provided an "excellent opportunity to study the effects of arsenic exposure on a homogeneous, stable population with a predominantly 'Mormon lifestyle'" (Southwick et al., 1981). The exposed communities of Hinckley and Deseret had average arsenic concentrations of 0.18 mg/l and 0.27 mg/l, respectively (based upon monthly water samples taken between May 1976 to May 1977). The control community of Delta had average arsenic concentrations of 0.02 mg/l. All drinking water in the study communities came from wells and the predominant species of arsenic was reported as the pentavalent form (85 percent).

TABLE 7-18. LANE COUNTY WATER ARSENIC LEVELS 1974-1978

Geographic region	Arsenic, ppb			1970 Population served by water districts	
	mean*	range	no. of samples	number	% of area residents
Rural	16.5+	0-2150	491	29,470	44
North	7.6	0-24	29	3,644	39
East	11.8	0-70	80	7,383	49
South	33.0	0-2150	186	12,713	59
Midwest	9.7	0-107	159	1,260	10
Coast	4.7	<1-13	37	4,470	54
Urban	4.8	0-860	67	135,260	91
NE Springfield	3.9	0-8	14	16,235	92
SW Springfield	6.0	0-30	4	18,460	96
S Eugene	5.3	0-750	10	29.230	88
W Eugene	6.5-	0-860	20	16,470	96
NW Eugene	3.8	<1-8	6	20,730	85
N Eugene	3.8	0-8	8	11,690	81
Core Eugene	4.0	<1-8	5	22,445	100
Lane County	8.6	0-2150	558	164,730	76

*Values with terminal digit of 5 indicate direction from which rounded, e.g., 0.51 - 0.5+, 0.49 - 0.5.

Source: Morton et al. (1976).

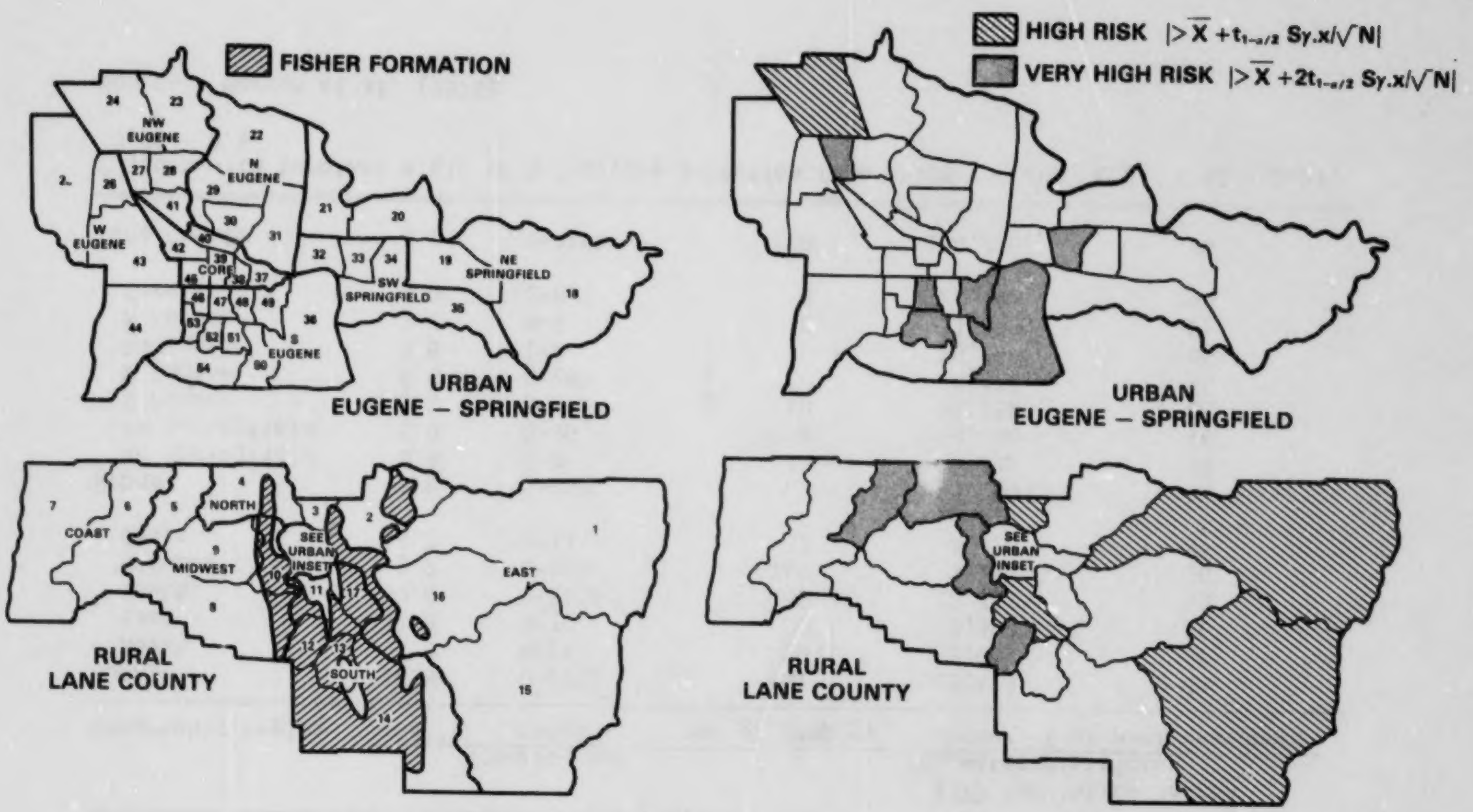

FISHER FORMATION
HIGH RISK |> X̄ + t1-α/2 Sy.x/√N|
VERY HIGH RISK |> X̄ + 2t1-α/2 Sy.x/√N|
NW EUGENE
N EUGENE
W EUGENE
CORE
S EUGENE
SW SPRINGFIELD
NE SPRINGFIELD
URBAN EUGENE – SPRINGFIELD
URBAN EUGENE – SPRINGFIELD
COAST
NORTH
MIDWEST
SEE URBAN INSET
EAST
SOUTH
RURAL LANE COUNTY
SEE URBAN INSET
RURAL LANE COUNTY

Figure 7-1. Comparison of census tracts experiencing exposure to the Fisher formation and exhibiting high skin cancer occurrence.

Source: Morton et al. (1975).

All study participants from the exposed communities were required to be five years of age or older and to have been residents of Hinckley or Deseret for at least the previous five years. Control participants were selected at random from age and sex categories matched to the exposed participants. Control participants were required to have lived their entire lives either in Delta or communities where arsenic in drinking water did not exceed the national standard of 0.05 mg/l.

Physical examinations were conducted to detect signs and symptoms associated with chronic arsenic poisoning. A total of 250 people participated in these examinations (145 exposed, 105 control); not all of these participated in each part of the examination, however. No explanation was provided by the authors as to the differences in participation rate for different parts of the examination. Urine and hair samples were collected from 94 and 74 percent of the participants, respectively. Dermatological examinations were conducted on 249 individuals. Neurological and hematological examinations were also conducted and are discussed in Sections 5.2.1 and 5.2.4, respectively. In addition, the incidence of cancer and vascular diseases in the study population was compared to other counties in the state of Utah.

The study results showed a clear relationship between the amount of arsenic consumed in drinking water and the amount measured in hair and urine. Differences between exposed and control populations were statistically significant.

Of the 249 participants examined for dermatological signs of arsenic toxicity (palmar and plantar keratosis and hyperkeratosis, tumors, diffuse pigmentation, arterial insufficiency), only 12 showed such signs and no participant had more than one sign. The 12 individuals were not clustered among the more heavily exposed, and when the dermatological signs were regressed against annual arsenic dose and the log of the dose, no significant associations were found.

Age-adjusted cancer incidence rates showed Hinckley to have a somewhat lower cancer incidence than Delta. Cancer death rates, 1956 to 1976, showed Hinckley to have the highest rate (138 per 100,000) when compared to 42 other Utah communities. Table 7-19 shows age-specific death rates for Utah and three Millard County communities. (Fillmore is the County seat; comparisons to Deseret were not made due to the community's small population). Between 1956 and 1976, 14 cancer deaths were reported for Hinckley. All of these deaths occurred in individuals 45 years or older and the cancers were types most frequently reported for Utah: lung, breast, large intestine, prostate, stomach, leukemia, kidney, uterus, bone and connective tissue. Hinckley had generally lower death rates for cardiac and vascular diseases than did the control community of Delta. The authors noted that no unusual death patterns likely to be associated with arsenic exposure were seen in Hinckley.

Certain weaknesses exist with this study, most notably, that of a small study population from which to derive meaningful statistical analyses. Furthermore, children and adults were not treated separately. In regard to sample size, the authors felt that the small sample size was somewhat compensated by the homogeneity and stability of the predominantly nonsmoking population. Data on food consumption were also missing which might have influenced urinary arsenic levels. This, in turn, may have resulted in overstating the strength of the dose-response relationship for urinary arsenic and arsenic in drinking water. It should also be noted that some participants were reported to have an average daily water consumption greater than 8ℓ, which seems very high, even

TABLE 7-19. AGE-SPECIFIC DEATH RATES FOR UTAH AND THREE MILLARD COUNTY COMMUNITIES (DEATHS/100,000/YEAR)

Community	Age Groups and (Number of Deaths)					
Characteristics	0-4	5-14	15-24	25-44	45-64	65+
CANCER						
State	7 (158)	6 (274)	6 (223)	19 (875)	149 (4749)	558 (7680)
Fillmore	0	14 (1)	0	17 (1)	196 (13)	706 (25)
Delta	0	0	22 (1)	61 (4)	189 (11)	986 (29)
Hinckley	0	0	0	0	390 (6)	1053 (8)
CARDIOVASCULAR						
State	1 (25)	0.4 (18)	1 (34)	18 (840)	275 (8788)	2079 (28587)
Fillmore	0	0	0	17 (1)	392 (26)	1638 (58)
Delta	0	0	0	30 (2)	344 (20)	1599 (47)
Hinckley	0	0	0	0	325 (5)	1316 (10)
CEREBROVASCULAR						
State	1 (24)	0.5 (23)	1 (36)	4 (201)	52 (1651)	797 (10965)
Fillmore	0	0	0	0	60 (4)	1017 (36)
Delta	0	0	0	0	34 (2)	1088 (32)
Hinckley	0	0	0	0	0	789 (6)
ARTERIOSCLEROSIS						
State	0	0.07 (3)	0.2 (7)	0.5 (21)	13 (406)	202 (2781)
Fillmore	0	0	0	0	15 (1)	311 (11)
Delta	0	0	0	0	17 (1)	272 (8)
Hinckley	0	0	0	0	65 (1)	395 (3)
AGE GROUP SIZE						
State	119,004	224,417	175,607	227,502	159,613	68,759
Fillmore	157	347	202	293	332	177
Delta	202	400	224	330	291	147
Hinckley	40	108	57	79	77	38

Death rates from 1956-1975 for cancer, cardiovascular disease, cerebrovascular disease, and arteriosclerosis, showing numbers of deaths and the average size of the age group.

Source: Southwick et al. (1981).

taking into account the hot summers. In reporting on the lack of arsenic-related effects in this study population compared to others (Tseng et al., 1968; Borgono and Greiber, 1972; Borgono et al., 1977; Zaldivar, 1977; Zaldivar and Guiller, 1977), the authors did note that populations in Taiwan and Antofagasta were exposed to considerably higher concentrations of arsenic in their drinking water.

Interestingly, the importance attached to the fact that no adverse effects were seen in this group of individuals exposed to drinking water arsenic levels four times that allowed by the Interim Primary Drinking Water Regulations (0.05 mg As/l) is somewhat compromised by the very characteristics of the population that make it useful for epidemiological study. The fact that this study population is so homogeneous and stable and, therefore, lends itself to a relatively well controlled statistical analysis, also makes it less useful in terms of being representative of the overall population of the U.S. For this reason, any generalizations that might be drawn from this study population are subject to limitations.

In addition to exposures via drinking water, food exposures and therapeutic exposures have also caused skin lesions and cancers. Hyperpigmentation and depigmentation of the skin were found to be common among the survivors of the Morinaga milk poisoning in 1955. A follow-up study conducted on the exposed children when they were 17-20 years of age showed the prevalence of lesions to be 15 percent (Yamashita et al., 1972). It is not known if any skin cancers have developed.

Hutchinson (1888) first reported on a possible association of skin cancer with the use of arsenical medicines. Prior to that, the association between these arsenical agents and keratotic lesions had been recognized. In his classic paper, Hutchinson reported on 6 patients with case histories who exhibited the keratotic lesions associated with arsenical poisoning. He felt that the clinical series supported two principal conclusions:

1. Prolonged internal use of arsenic may seriously affect the nutrition of the skin and that use may produce warty or corn-like indurations.
2. Continued use of arsenic may result in a tendency for these "arsenic corns" to grow downward and pass into epithelial cancer.

Neubauer (1947) later compiled a series of reports on 143 cases of medicinal arsenical epitheliomas. He excluded five categories of cases reported in the literature to keep the series consistent with regard to diagnosis of case and history of arsenical use. Seventy-one percent of the patients being treated with arsenical medicine were patients suffering from skin diseases, especially psoriasis (54 percent). In contrast, only a small percentage of the cases reported came from patients treated with arsenic for various internal disorders. In nearly all cases the drugs used were inorganic and almost all were trivalent. The most commonly used arsenical was potassium arsenite. No externally applied arsenic-related skin cancer case was reported in the series.

The elapsed time from the beginning of administration of the arsenical drug to the beginning of the epitheliomatous growth was variable, but averaged 18 years regardless of the type of lesion. In cases with keratosis, the latent period to the onset of keratosis was about half the latent period to the onset of the epithelioma, i.e., about 9 years. Thirty-three percent of the patients were 40 or younger. Of the 143 patients, 13 had or developed miscellaneous cancers at other sites, but such cases were not reported systematically.

Fierz (1965) reported a follow-up study of patients treated with arsenic by a private practitioner. An accurate assessment of the total arsenic intake in terms of the amount of Fowler's Solution administered was available to the investigator from patient records. The follow-up examination was conducted under the auspices of a local polyclinic. Fourteen hundred fifty patients were identified as having received arsenic treatment, and invitations were mailed for them to come for a free follow-up medical examination. During the period March 1963 to April 1964, 262 patients presented themselves for examination. Two hundred eighty patients were not located while 100 patients actively refused to participate. Only patients under 65 years of age were invited to participate in the study. The author admits that the patients reporting for examination were not a representative sample. In fact, he categorizes them into three groups which range the spectrum of likely biases. There were patients satisfied with the results of the arsenic treatment and wishing to express thanks, patients in whom disturbing side effects were occurring, and finally patients who were still suffering from the initial disease and who were eager to get a consultation.

Arsenic treatment was prescribed for individuals suffering primarily from three main skin diseases: psoriasis (64 patients), neurodermatitis (62), and chronic eczema (72). In addition, treatment was also prescribed for 64 patients suffering from assorted skin diseases other than those listed above.

Fierz noted that the arsenic treatment showed good success. Of the 64 cases of psoriasis, 55 reported a favorable effect while taking the drops. Forty-eight of 62 patients with neurodermatitis reported a favorable effect. This effectiveness was the cause for the patients' reliance on the drug.

Upon examination, 106 of 262 patients (40.4 percent) reported hyperkeratoses, although frequently a detailed examination was necessary to find the changes. Hyperkeratoses were round, superficially verrucose papules, 1 to 3 mm in diameter. The number and the exact presentation of the hyperkeratoses varied from case to case.

In the series, 21 cases of skin cancer were found comprising 8 percent of the total subjects. As was in the case of hyperkeratosis, some variability was observed in the expression of the skin cancers. Multiple basal cell carcinoma was the most frequent histologic type observed, occurring in 48 percent of the cancer patients. The basal cell carcinomas appeared morphologically as polycyclic, sharply bounded erythemas with slight infiltration. In 13 of the 21 cancer cases, multiple carcinomas were observed, a much higher proportion than had been observed with other causes of skin cancer. Of the 21 patients with carcinomas, 16 showed distinctly developed arsenic warts on the palms and soles simultaneously with the skin tumors.

Both hyperkeratosis and skin cancer were found to vary with increasing arsenic intake. Above 400 ml of Fowler's solution, more than 50 percent of the patients studied showed hyperkeratosis. As the dose increased there was a corresponding increase in the simultaneous occurrence of hyperkeratosis of the palms and soles. Only 2/15 cases of hyperkeratosis with intakes of up to 250 ml had a simultaneous presentation, while 16/27 had a simultaneous presentation with intake of 250 to 500 ml.

As with the hyperkeratosis, skin cancer increased with increasing arsenic doses. Below 500 ml only basal cell carcinomas were found; spindle cell carcinomas were only found above that dose. The latency period for hyperkeratosis was at a minimum 2.5 years; skin cancer, however, had a minimum latency period of 6 years with a mean latency period of 14 years.

Cuzick et al. (1982) conducted a mortality analysis of a cohort of 478 patients given Fowler's solution to determine if persons taking the solution were at an increased risk of internal malignancies. The cohort consisted of 213 males (45 percent) and 265 females (55 percent) who took Fowler's solution for various lenghts of time ranging from 2 weeks to 12 years (mean duration, 8.92 months) during the period of 1945-1969. Follow-up was until January 1, 1980; the mean follow-up time for the entire cohort was 20.3 years. The median cumulative dose of arsenic was reported as 448 mg, with most patients consuming arsenic at a rate of about 250 mg/month.

Within the defined risk period, 139 patients died, 265 patients were known to be alive and at risk, 29 had passed 85 years of age, 19 had emigrated, and 26 could not be traced. Because of the unreliability of the expected values in the open-ended group beyond 85 years of age, deaths and person-years at risk after age 85 were ignored. Persons who emigrated were censored as of the date of emigration.

A subset of 142 of the cohort were physically examined during 1969-70, and the presence of arsenical keratoses, hyperpigmentation, and skin cancer was recorded. No signs of internal malignancy were clinically apparent at the time of examination. This subset was chosen because the individuals were known to be alive and were able to be traced.

The results showed that risk ratios were similar for both males and females. No signs of internal malignancy were clinically apparent in the patients given physical examinations, and mortality was not found to be significantly increased for any internal malignancies in the cohort, although it was somewhat elevated for death from bladder cancer (observed = 3, expected = 1.19, P = 0.12). The ratio of observed-to-expected deaths from all neoplasms, from cancer of the digestive organs, from cancer of the respiratory organs, and from all causes was analyzed by cumulative dose level. No trend by cumulative dose level was found; however, the risk ratios were low for deaths from all neoplasms, digestive cancers, and respiratory cancers in the lowest dose group, with the risks for deaths from all neoplasms being significantly ($p < 0.05$) lower than expected. A significant positive trend in the ratio of observed-to-expected mortality for all neoplasms was found for dose in the period 5-9 years from first exposure, but this trend was not found in other time intervals since first exposure.

Of the subset of 142 patients examined for skin manifestation of arsenicism, their overall mortality was consistent with the remainder of the cohort. Within this group, 45 percent had keratosis, 14 percent had hyperpigmentation, and 11 percent were found to have skin cancer. The authors did not indicate what the expected proportions of patients with these signs would be. Some patients showed 2 or all 3 signs of arsenicism, and 49 percent showed at least one of the signs. The appearance of signs was dose- and age-related. Patients with signs had higher median doses (672 mg) than those without any signs (448 mg), and this was significant ($P < 0.001$) by the Wilcoxon rank-sum test. Forty percent of patients with signs had cumulative doses above 1000 mg, whereas only 22 percent of the patients without arsenical signs had cumulative doses above 1000 mg. On the average, patients with signs were 7 years older and had received their first exposure 6 calendar years earlier than those without signs. Within the group with exposures > 1000 mg, those with signs were, again, an average of 7 years older, and they had their first exposure 3 years earlier. It is interesting to note that all seven of the subsequent deaths from internal malignancy in this subgroup had been identified as showing signs of arsenicism. No deaths from cancer occurred in the group without physical signs.

In summary, this study provided little evidence that ingestion of arsenic is related to a risk of mortality from internal malignancies. Non-fatal skin cancer was found, however, to be related as a function of dose to ingestion of Fowler's solution. The authors indicated that perhaps persons who demonstrate signs of arsenicism following arsenic exposure retain arsenic longer than those who do not show signs, which may suggest that persons who demonstrate signs of arsenicism are at a greater risk of internal malignancies. This was suggested by the fact that all of the deaths from internal malignancies in the subset of individuals that was examined for signs of arsenicism occurred in individuals who demonstrated such signs.

Knoth (1966) also reported on two patients treated with arsenical medicinals and who developed skin cancer. Roth (1958) reported that of 47 autopsy cases of vintners with chronic arsenic intoxication, 13 had skin tumors.

Gilbert et al. (1983) conducted a study on wood treaters in Hawaii who were occupationally exposed to arsenic (see Section 7.1.2.1 for complete discussion of study). The authors found no evidence of risk of skin cancer, but inadequacies in the study design and the small sample size limit the ability to draw valid conclusions from the data.

7.1.2.3 Other Cancers--Whereas there are many studies suggesting that there is an association between inhalation exposure and respiratory cancer and oral exposure and skin cancer, respectively, there are no consistent data with regard to cancer in internal organs.

Reymann et al. (1978) have investigated the relationship between the intake of arsenic for medicinal purposes and subsequent internal neoplasms. Study subjects were identified by examining the files of a dermatology clinic in Denmark for the years 1930 to 1939. Two rosters of study subjects were generated: 1) persons treated with arsenic for multiple basal cell carcinoma, Bowen's disease, psoriasis, verruca planus, and lichen planus, and 2) persons with keratoses from the first roster plus 30 other persons identified as having keratoses. The first roster of 413 persons comprised the basic study population; 24 persons were excluded for various reasons, resulting in a final study population of 389 persons. The malignancy histories of both populations were traced through the years 1943-1974 in the Danish Cancer Registry. For each person a length-of-observation period was determined and an expected number of internal malignant neoplasms was determined using the former data.

In the main study population, 41 cases were observed versus 44.6 cases expected. Therefore, no increased incidence of internal malignancies was noted in the arsenic-treated patients. However, examination of the data by individual skin disease categories showed that women with multiple basal cell carcinoma had a significantly higher incidence of internal cancer. The same trend, although not significant, was observed in female patients exhibiting verruca planus. Next, an analysis was made of a possible dose-response relationship using duration of treatment as the exposure variable. The categories were low, medium and high, based on <1 month, 1-3 month, and >3 month duration of standard dose administration (6-8 mg As_2O_3). No relationship between dose and internal organ cancer was noted for the total population or by sex. In addition, the form of arsenic administered did not seem to affect the incidence of internal cancer, nor was any effect noted due to period of observation. Reymann et al. did not provide a definition of the term "internal cancer." It is presumed that the authors included lung cancer in their definition. If so, the authors did not provide any adjustment for smoking. Regardless, the sample size of 389 persons is probably too small to detect an excess in the incidence of internal cancer, even if lung cancers were excluded.

The keratosis group was not analyzed in the same way. Eight of the 19 men with keratosis died before 1974. Four of these were due to cancer of the internal organs. Expected deaths were 1.9. Similarly, 5 deaths due to cancer of the internal organs were observed in the 34 women with keratosis compared with 3.3 expected. Together there were 9 deaths due to cancer of the internal organs compared to 5.2 expected. Although the figure is almost doubled, it is still not significant. On the average, the patients with keratosis received higher doses of arsenic than the other patients.

Hemangioendothelioma and reticulosarcoma of the liver has been reported by Roth (1958) to occur among German vintners exposed occupationally to arsenic. The same type of malignancy occurrence has also been reported in isolated cases by other authors (Pershagen and Vahter, 1979). Higgins et al. (1982) reported increased mortality for cirrhosis of the liver and urinary cancer in workers at the Anaconda Montana smelter.

More recently, Falk and coworkers (1981a) described a nationwide (U.S.) review of deaths from hepatic angiosarcoma during the years 1964-1974. Of 168 confirmed cases included for survey, a group of seven cases with a history of prolonged use of Fowler's solution was found. Taken with the data of Roth, chronic arsenic exposure, mainly via ingestion, appears to be associated with this very rare liver cancer.

In a review of four cases of hepatic angiosarcoma, Falk et al. (1981b) determined that one case, that of a child, involved environmental arsenic exposure in a copper-smelting community in Arizona. Since this child had a history of pica, the usual exposure routes were augmented by further arsenic intake.

Roat et al. (1982) reported a case of hepatic angiosarcoma in a subject who, 33 years earlier, had ingested Fowler's solution for the relatively short period of 6 months. The pre-existence of skin cancer in this subject, from age 25, of the type associated with arsenic exposure, was further support of an arsenic association.

Knoth (1966) reported on two patients who had been treated with arsenical medicinals and developed tumors at sites other than the skin or lung. One of the patients was a 61-year old woman who had been treated with an arsenical medicinal and developed mammary carcinoma. The other case was that of a 53-year old male who had been treated with an arsenical medicinal for cirrhosis vulgaris and developed a reticulosarcoma of the glans penis.

In the studies by Ott et al. (1974), an increased proportionate mortality due to malignant neoplasms of lymphatic and hematopoietic tissues was found. Axelson et al. (1978) found an increased risk of leukemia and myeloma in a case-control study of workers exposed at a smelter.

7.2 ANIMAL STUDIES

While arsenic carcinogenicity in test animals has not been observed in most studies, some recent reports have noted positive results. The literature on experimental inorganic arsenic carcinogenesis, as summarized in Table 7-20 and as reviewed by scientific bodies (NAS, 1977; NIOSH, 1975; IARC, 1973 and 1980) and individuals (Sunderman, 1976; Wildenberg, 1978; Pershagen and Vahter, 1979) supports this conclusion.

In light of the presently recognized anomalous metabolizing of inorganic arsenic by rats, studies which used rats as the experimental subjects should be viewed cautiously. Studies on other animal models have generally resulted in negative findings. A few of these studies are discussed below, but most have been summarized in Table 7-20.

In a study by Baroni et al. (1963), Swiss mice were given either arsenic trioxide dissolved in drinking water (concentration of 100 mg/ℓ) ad libitum for the duration of the 70-week experiment; or, sodium arsenate in a concentration of 15.8 gm/ℓ in a 2.5 percent solution of Tween 60 in water, applied twice weekly for the duration of the experiment. Each compound was tested alone, in combination with skin applications of croton oil (to test for initiating action), and after initiation with a single skin application of 7,12-dimethylbenz(a)anthracene or with administration of urethan by stomach tube

TABLE 7-20. SUMMARY TABLE OF EXPERIMENTAL STUDIES OF ARSENIC CARCINOGENESIS

Route	Species (Strain)	Compound	Vehicle	Results*	Reference	Remarks
Oral	Mice (C57B16)	As_2O_3	Tap water or 12% aqueous ethanol	-	Hueper and Payne, 1962	Shortened life span of treated (aq. ethanol) vs. control.
Oral	Mice (Swiss)	As_2O_3	Drinking water	-	Baroni et al., 1963	
Oral	Mice (Swiss)	$NaAsO_2$	Drinking water	-	Kanisawa and Schroeder, 1967	Shortened life span, low dose.
Oral	Mice (NMRI)	As_2O_3	Drug (Psor-Intern) or Fowler's solution	+(?)	Knoth, 1966	Significance cannot be determined, uncomplete reporting by author.
Oral	Mice (C3H/St, female)	$NaAsO_2$	Drinking water	-	Schrauzer and Ishmael, 1974	
Oral	Rats (not specified)	$Pb_3(AsO_4)_2$ $Ca_3(AsO_4)_2$	Not specified	-	Fairhall and Miller 1941	Poor survival in treated. As reviewed by IARC (Vol. 23, 1980)
Oral	Rats (Bethesda Blacks)	As_2O_3	Tap water or aqueous ethanol (12%)	-	Hueper and Payne, 1962	
Oral	Rats (Osborne-Mendel)	$NaAsO_2$ Na_3AsO_4	Diet	-	Byron et al., 1967	Reduced survival at highest dose.
Oral	Rats (Long Evans)	$NaAsO_2$	Drinking water	-	Kanisawa and Schroeder, 1969	Low dose.
Oral	Rats (Wistar)	$Pb_3(AsO_4)_2$ Na_3AsO_4 $Pb_3(AsO_4)_2$ + NDEA Na_3AsO_4 + NDEA	Diet Diet Diet Diet	- - - -	Kroes et al., 1974	High mortality and 2 tumors (incidence not reported observed in lead arsenate group).

TABLE 7-20. (continued)

Route	Species (Strain)	Compound	Vehicle	Results*	Reference	Remarks
Oral	Dog	$NaAsO_2$ or Na_3AsO_4	Diet	-	Byron et al., 1967	Weight loss, early mortality, short duration of experiment.
Inhalation	Mice (not specified)	$NaAsO_2$	Aqueous aerosol	-	Berteau et al., 1978	Reported in an abstract form, high incidence in controls.
Intratracheal instillation	Rats (Wistar King) male	(1) As_2O_3 (2) Copper ore (3.95% Arsenic)	Not specified	- -	Ishinishi et al., 1977	
		(3) Flue dust		-		One tumor observed in treated, not significant.
Intratracheal instillation	Rats (Wistar King) male	As_2O_3	Aqueous solution	-	Ishinishi et al., 1976	One tumor observed in treated, not significant.
Intratracheal instillation	Rats (BDIX)	$Ca_3(AsO_4)_2$	Bordeaux mixture (contains $CuSO_4$ and $Ca(OH)_2$)	+	Ivankovic et al., 1979	Results cannot be attributed to arsenic alone.
Intratracheal instillation	Golden Hamsters (males)	As_2O_3 ± BP	Carbon carrier + dilute H_2SO_4	+	Pershagen et al., 1983	Adenomas significantly greater than controls ($p < 0.01$). Carcinomas in 3 treated animals vs. 0 in controls.
Intratracheal instillation	Golden Hamsters (female)	As_2O_3	Phosphate buffer	+	Ishinishi and Yamamoto, 1983	As_2O_3 was tumorigenic in a dose-response fashion.

TABLE 7-20. (continued)

Route	Species (Strain)	Compound	Vehicle	Results*	Reference	Remarks
Intratracheal instillation	Rats	Ore dust containing As at two levels of content	Not specified	+	Chung and Liu, 1982	Possible confounding effects of other elements present, unknown. No apparent differences in lung cancer with different As levels in dust.
Skin painting (application)	Mice (not specified)	$KAsO_2$ and As_2O_3	Ethanol	-	Leitch and Kennaway, 1922	Neubauer (1947) failed to confirm this observation.
Skin painting (application)	Mice (S)	$KAsO_2$ followed by croton oil in acetone	Methanol	-	Salaman and Roe, 1956	As reviewed by IARC (Vol. 23, 1980).
Skin painting (application)	Mice (Rockland all-purpose)	(1) $KAsO_2$ followed by croton oil in benzene	80% ethanol	-	Boutwell, 1963	As reviewed by IARC (Vol. 23, 1980).
Skin painting (application)	Mice (Rockland all-purpose)	(2) DMBA followed by $KAsO_2$ in 80% ethanol	acetone	Not carcinogenic	Boutwell, 1963	
Skin painting (application)	Mice (Swiss)	(1) Na_3AsO_4	Water containing Tween 60	-	Baroni et al., 1963	
		(2) Na_3AsO_4 followed by croton oil and DMBA	Water containing Tween 60	No promotional activity observed		
Subcutaneous injection	Mice (Swiss) female Progeny	Na_3AsO_4 Na_3AsO_4	Water Water	+ +	Osswald and Goerttler, 1971	

TABLE 7-20. (continued)

Route	Species (Strain)	Compound	Vehicle	Results*	Reference	Remarks
Subcutaneous implant	Rats (random-bred) male albino	$Ca_3(AsO_4)_2$	Paraffin pellets	-	Arkhipov, 1968	As reviewed by IARC (Vol. 23, 1980).
Subcutaneous injection	Rats (random-bred) male albino	$Ca_3(AsO_4)_2$	Sunflower oil	-	Arkhipov, 1968	
Subcutaneous injection (pre-natal + post-natal)	Mice (CFLP)	As_2O_3	Aqueous solution	+	Rudnai and Borzsony, 1981	Histological description of tumors poor.
Intravenous injection	Mice (Swiss) female	Na_3AsO_4	Water	+	Osswald and Goerttler, 1971	
Intramuscullary injection	Rats (Osborne-Mendel)	Arsenic	Metallic	-	Hueper, 1954	
Intramuscullary injection	Rabbits	Arsenic	Metallic	-	Hueper, 1954	
Oral	Mice (Swiss)	As_2O_3 and croton oil, DMBA or urethan	Drinking water	-	Baroni et al., 1963	
Oral	Mice (STS)	Arsanilic acid or $KAsO_2$ followed by DMBA (skin) then croton oil (skin) in benzene	Diet	-	Boutwell, 1963	As reviewed by IARC (Vol. 23, 1980).

(*) + statistically significant excess tumors observed over controls

- no statistically significant excess tumors of treated vs. control or no tumors observed (to test for promoting action). All tests failed to show any carcinogenic activity of the two compounds under the given experimental conditions.

Kanisawa and Schroeder (1969) treated Swiss mice with sodium arsenite (equivalent to 5 μg/ml As) in drinking water for the 30-month duration of their study. In the treated animals, the authors noted 6 malignant and 11 combined malignant and benign tumors out of 103 animals versus 15 malignant and 50 combined malignant and benign tumors out of 170 control animals; thus, the results were negative for showing any carcinogenic effect of sodium arsenite in this study.

In a study of Leitch and Kennaway (1922; as reported in IARC, 1980), 100 mice were given skin applications of a solution of potassium arsenite in ethanol containing 1.8 percent arsenic trioxide (later reduced to 0.12 percent due to a high death rate), thrice weekly for 3 months. Of the 33 mice that lived for 3 months, only one developed a metastasizing squamous cell carcinoma.

In certain animal systems, positive carcinogenic responses have been reported, however. For example, Osswald and Goerttler (1971) exposed pregnant Swiss mice to daily parenteral dosing of sodium arsenate (0.5 mg/kg, solution of 0.005 percent arsenate salt) for a total of 20 injections. Part of the offspring groups received 20 injections of the same level subcutaneously at weekly intervals. Leukemia or lymphoma was seen in 46 percent of the mothers (11/24) at the end of 2 years versus none in the controls. Of the treated offspring, 41 percent (17/41) of the males and about half of the females (24/50) developed leukemia versus only 3 of 55 male and female control offspring (approximately 6 percent). IARC (1973) has criticized this study for the absence of exposure of controls to the appropriate vehicle solution.

Knoth (1966) noted a significant frequency of tumors in 30 mice exposed to Fowler's solution orally (one drop/week, 20 weeks, approximately 5.3 mg As total),including adenocarcinomas of the skin, lung, and lymph nodes. The absence of experimental details makes critical assessment of this study difficult.

Some recent animal studies have employed different exposure conditions than have been employed in the past. Berteau et al. (1978) have exposed a tumor-susceptible strain of female mice to a respirable aerosol of inorganic arsenic (1 percent aqueous solution of sodium meta-arsenite, 20 to 40 minutes daily, 5 days/week, 55 weeks total). The 30 exposed mice showed neither gross nor histological evidence of neoplasia.

In a two-part study, Ishinishi et al. (1977) examined the carcinogenic and co-carcinogenic effects of various arsenic compounds on male Wistar-King rats. In the first part of the study, arsenic trioxide, an arsenic-containing copper ore (containing 3.95 percent arsenic) or metal refinery flue dust (containing 10.5 percent arsenic) were administered to 51 rats via 15 weekly intratracheal instillations. The rats were observed over their lifespan. Of the 25 surviving rats, one adenocarcinoma was seen in the group receiving flue dust. One lung metastasis from osteosarcoma of the femur and one adenoma was reported in the group exposed to the copper ore. No malignant tumors were reported in the group receiving arsenic trioxide; however, one adenoma was reported for this group. All groups displayed squamous cell metaplasia in the airway and osteometaplasia in the alveolus of the lung.

In the second part of the study, 87 rats were instilled with each of the above compounds suspended in a saline solution containing benzo[a]pyrene (B[a]P) or with B[a]P, alone. Control rats (23) were instilled with the saline solution. Of the 34 surviving exposed rats, one adenocarcinoma was seen in the group receiving B[a]P plus copper ore. All exposed groups had squamous cell carcinomas of the lung. Of particular interest to the authors, was the noted co-carcinogenic effect of arsenic trioxide with B[a]P; rats in this group exhibited a 43 percent incidence rate (3/7) for squamous cell carcinomas. This compared with a 14 percent (1/7) incidence rate in animals given B[a]P, alone. No benign or malignant tumors were seen in the 7 surviving control rats. Again, squamous cell metaplasia or osteometaplasia were seen in all groups.

The results indicated a positive interaction between arsenic trioxide and B[a]P; however, the authors noted that the numbers of surviving animals were too small to permit drawing any firm conclusions from the study.

In another study, Ishinishi et al. (1980) gave 30 male adult Wistar rats intratracheal instillations of arsenic trioxide in suspension for 15 weeks. Of the 19 rats that survived, only one malignant squamous cell carcinoma was observed over lifetime. No tumors were found in the controls.

Ivankovic and co-workers (1979) exposed rats, via intratracheal instillation, to a pesticide mixture corresponding to that used in the past for vineyard treatment and consisting of a mixture of copper (II) sulfate, calcium hydroxide and calcium arsenate. Of 25 rats exposed to approximately 0.07 mg arsenic, 10 died from lung necrosis or pneumonia. In the survivors, 9 animals (60 percent) showed multi-focal bronchogenic adenocarcinomas and bronchiolar/alveolar cell carcinomas.

This study appears to offer experimental evidence that the vineyard pesticide mixture, employed as such, could have been carcinogenic to vine dressers working with the material. One difficulty with this study is an ambiguity regarding its full significance for the general issue of arsenic-inducing carcinogenic effects by itself. Clearly, the high mortality rate, 40 percent, and the known toxicity of Bordeau mixture (copper sulfate plus calcium hydroxide) to animals (Pimentel and Marques, 1969) and man (Villar, 1974, Pimentel and Marques, 1969) suggest carcinogenesis; however, it is impossible to clearly ascribe such activity to arsenic alone given the presence of other compounds within the mixture. In their studies, Ivankovic et al. (1979) did not include animal groups exposed to calcium arsenate alone or to Bordeau mixture alone.

Pershagen et al. (1983, in press) have studied the pulmonary carcinogenicity of arsenic trioxide alone and in combination with benzo(a)pyrene in male golden hamsters given 15 weekly intratracheal instillations of 3 mg As/kg and/or 6 mg/kg B[a]P. In this study, a carbon dust carrier with dilute sulfuric acid was employed to enhance retention of arsenic. Carcinomas of larynx, trachea, bronchi and lungs were found in 3 animals given just arsenic versus a zero response in controls. The incidence of adenomas, papillomas and adenomatoid lesions was higher in arsenic versus control groups ($p < 0.01$). While the carcinomas in the arsenic group did not reach statistical significance, the significantly higher incidence of the adenomas, papillomas, and adenomatoid lesions support the authors' conclusion that the carcinomas were not a chance finding. The use of the carrier to enhance lung retention of arsenic appears to be the key to the results in this study. By itself, the oxide of arsenic is rather rapidly cleared from the lung (see below).

Rudnai and Borzsony (1981) reported that a combination of pre- and postnatal exposure of CFLP mice to injected arsenic trioxide (1.2 µg As/g, days 15-18 of gestation; 5 µg As/animal for 3 days post-birth) resulted in a 63 percent lung tumor incidence at 1 year of age versus approximately 18 percent in controls. In this study, a detailed histological description of the tumors was not given, but it can be assumed that both adenomas and adenocarcinomas were observed.

Chung and Liu (1982) reported that the repeated intratracheal instillation of ore dust containing arsenic in rats resulted in a lung cancer incidence rate of approxiamtely 15 percent (6/41) over a period of 3 years, while controls (N=18) showed no lung tumors. There was extensive metastasis in one animal. In this study, two compositions of metal content in ores were used, with the arsenic varying by 8-fold. The lung cancer rate for the two forms of ore dust were comparable, despite the 8-fold difference in arsenic, i.e., there was no simple dose response. On the other hand, the ores contained variable amounts of iron and lead, which may have affected both the dose response and, possibly, the actual incidence of the lung cancers.

Ishinishi and Yamamoto (1983) exposed female golden hamsters to arsenic trioxide by intratracheal instillation of a suspension in phosphate buffer, 15 times weekly, for 4 months, at a total dose of 5.3 mg As. A second group was given a total dose of 3.8 mg As. All animals were followed over their entire life span. The incidence of lung tumors (adenomas) for the combined exposure groups was approximately 17 percent versus no tumors in surviving controls. While the sample size was small, there was the suggestion of a dose response across the two exposure levels (3/10 for the higher dose; 1/10 for the lower dose). These data suggest that arsenic trioxide is tumorigenic in the golden hamster and may be carcinogenic.

Recently, both Inamasu et al. (1982) and Pershagen et al. (1982) have studied the effects of intratracheal instillation of calcium arsenate (see Section 4.1.1 for complete discussion of studies). Inamasu et al. gave single intratracheal instillations of arsenic trioxide or calcium arsenate to male Wistar rats. Pershagen et al. instilled male Syrian golden hamsters with four weekly suspensions of arsenic trioxide, arsenic trisulfide and calcium arsenate. The results of both studies showed that arsenic trioxide was rapidly cleared from the lungs, whereas calcium arsenate was slowly eliminated. The differences in clearance appeared to be related to solubility, with the less soluble calcium arsenate exhibiting the slowest clearance.

These recent findings might help to explain the differences noted above in the earlier studies of Ishinishi et al. (1977; 1980) and Ivankovic et al. (1979). In regard to the Ivankovic study, it may be that the calcium arsenate, of itself, contributed to the high mortality rate observed in the exposed rats.

Schrauzer and co-workers (Schrauzer and Ishmael, 1974; Schrauzer et al., 1977; Schrauzer et al., 1978) have reported experimental results using oral arsenic and the tumorigenic effect of the agent on spontaneous mammary adenocarcinomas in an inbred strain of mice (C_3H/St Mice). These workers noted that while arsenic retards the overall incidence of tumor formation (10 or 80 ppm As), it stimulates the growth of tumors that otherwise occur. When these animals were exposed to only 2 ppm As (arsenite) in drinking water, compared to levels of 10 or 80 ppm, there was no effect on frequency of tumors, although the same enhanced tumor growth was seen as before with levels of 10 or 80 ppm As (Schrauzer et al., 1978). Furthermore, a higher incidence of multiple tumors of the mammary gland was observed, as was the abolishing of the anticarcinogenic effect of selenium in this system when both elements were given together.

In support of the positive responses noted in the above animal studies, Dipaolo and Casto (1979) reported the transformation of cultured Syrian hamster embryo cells by direct exposure to sodium arsenate (NA_2HAsO_4). In their assay, 300 cells from secondary cultures of Syrian hamster embryo cells (HEC) were plated in complete medium with 20 percent serum in 50-mm plastic Petri dishes along with 6×10^4 HEC cells which had been irradiated as confluent monolayer cultures. Sodium arsenate at concentrations of 0 μg/ml (control), 2.5 μg/ml and 5.0 μg/ml in complete medium (with a final concentration of acetone less than 0.02 percent) were added 24 hours later. Dishes were fixed and stained nine days after seeding. Colonies were scored blind by two observers.

For the two sodium arsenate concentrations, the authors reported that the number of transformations were 1.16 and 2.08/dish, respectively, or 1.74 and 4.13 percent, respectively, on the basis of transformed colonies relative to total colonies scored. The authors also reported that typical colonies scored as transformed after direct treatment with a metal carcinogen were identical to those obtained with other chemical carcinogens. In addition, the authors claimed (data not shown) that when 2.5 μg benz(a)pyrene per ml of medium were used, the transformation frequency, on a colony basis, was 4 to 6 percent.

7.3 QUANTITATIVE CARCINOGEN RISK ESTIMATES

7.3.1 Introduction

This quantitative section deals with the unit risk for arsenic in air and water and the potency of arsenic relative to other carcinogens that the Carcinogen Assessment Group (CAG) of the U.S. Environmental Protection Agency has evaluated. The unit risk estimate for an air pollutant is defined as the lifetime cancer risk occurring in a population in which all individuals are exposed continuously from birth throughout their lifetimes to a concentration of 1 μg/m^3 of the agent in the air they breathe. The unit risk estimate for water is defined similarly, but with a water concentration of 1 μg/ℓ. Unit risk estimates are used for two purposes: (1) to compare several agents with each other in terms of carcinogenic potency, and (2) to give a crude indication of the human health risks that might be associated with exposure to these agents, if the actual exposures are known.

The data used for quantitative estimates can be of two types: (1) lifetime animal studies, and (2) human studies where cancer risk has been associated with exposure to the agent. It is assumed, unless evidence exists to the contrary, that if a carcinogenic response occurs at the dose levels used in a study, then responses at all lower doses will occur with an incidence that can be determined by an appropriate extrapolation model.

There is no solid scientific basis for any mathematical extrapolation model that relates carcinogen exposure to cancer risks at the extremely low concentrations which must be dealt with in evaluating environmental hazards. For practical reasons, such low levels of risk cannot be measured directly either by animal experiments or by epidemiologic studies. It is necessary, therefore, to depend on current knowledge of the mechanisms of carcinogenesis for guidance as to the correct risk model to use.

At the present time, the dominant view is that most cancer-causing agents also cause irreversible damage to DNA--a position supported by the fact that a large proportion of agents that cause cancer are also mutagenic. There is reason to expect that the quantal type of biological response, which is characteristic of mutagenesis, is associated with a linear non-threshold dose-response relationship. Indeed, there is substantial evidence from mutagenesis studies with both ionizing radiation and a wide variety of chemicals that this type of dose-response model is the appropriate one to use in estimating cancer risks from environmental exposures. This is particularly true at the lower end of the dose-response curve. At higher doses, there can be an upward curvature, probably reflecting the effects of multistage processes on the mutagenic response. The linear non-threshold dose-response relationship is also consistent with the relatively few epidemiologic studies of cancer responses to specific agents that contain enough information to make the evaluation possible (e.g., radiation-induced leukemia, breast and thyroid cancer, skin cancer induced by arsenic in drinking water, liver cancer induced by aflatoxins in the diet). There is also some evidence from animal experiments that is consistent with the linear non-threshold model (e.g., the initiation stage of the two-stage carcinogenesis model in rat liver and mouse skin).

Because of these factors, the linear non-threshold model is considered to be a viable possibility for the true dose-response relationship and, unless direct evidence to the contrary is presented, it is used as the primary basis for risk extrapolation at low levels of exposure.

The quantitative aspect of carcinogen risk assessment is included here because it may be of use in setting regulatory priorities, evaluating the adequacy of technology-based controls, and other aspects of the regulatory decision-making process. However, the imprecision of presently available technology for estimating cancer risks to humans at low levels of exposure should be recognized. At best, the linear extrapolation model used here provides a rough but plausible estimate of the upper limit of risk--that is, with this model it is not likely that the true risk would be much more than the estimated risk, but it could be considerably lower. The risk estimates presented below should not be regarded, therefore, as accurate representations of true cancer risks even when the exposures involved are accurately defined. The estimates presented may, however, be factored into regulatory decisions to the extent that the concept of upper-risk limits is found to be useful.

7.3.2 Unit Risk for Air

7.3.2.1 Methodology for Quantitative Risk Estimates--The methodologies used to arrive at quantitative estimates of risk must be capable of being implemented using the data available in existing epidemiologic studies of exposure to airborne arsenic. In order to extrapolate from the exposure levels and temporal exposure patterns in these studies to those for which risk estimates are required, it will be assumed that the age-specific mortality rate of respiratory cancer per year per 100,000 persons for a particular 5-year age interval, i, can be represented using either of two models:

$$a_i(D) = a_i[1 + a'D^k] \qquad (1)$$

(a relative or multiplicative risk model), or

$$a_i(D) = a_i + 100{,}000a'D^k \qquad (2)$$

(an absolute or additive risk model). With either model, a_i is the age-specific mortality rate per year of respiratory cancer in a control population not exposed to arsenic, a' is a parameter representing the potential of airborne arsenic to cause respiratory cancer, and D is some measure of the exposure to arsenic up to the ith age interval. For example, D might be the cumulative dose in $\mu g/m^3$ years, the cumulative dose neglecting exposure during the last 10 years prior to the ith age interval, or the average dose in $\mu g/m^3$ over some time period prior to the ith age interval. The forms to be used for D will be constrained by the manner in which dose was treated in each individual epidemiologic study. The parameter k determines the shape of the dose-response curve. Attention will be given particularly to the values k = 1 and k = 2. If k = 1, the age-specific incidence rates vary linearly with the dose level (a linear model), and if k = 2 they vary quadratically. At low exposures the extra lifetime probability of respiratory cancer mortality will vary correspondingly (e.g., linearly for k = 1 and quadratically for k = 2).

The dose-response data available in the epidemiologic studies for estimating the parameters in these models consists primarily of a dose measure D_j for the jth exposure group, the person-years of observation Y_j, the observed number of respiratory cancer deaths O_j, and the number E_j of these deaths expected in a control population with the same sex and age distribution as the exposure group. The expected number E_j is calculated as

$$E_j = \sum_i Y_{ji} a_i / 100{,}000, \qquad (3)$$

where Y_{ji} is the number of person-years of observation in the ith age category and the jth exposure group ($Y_j = \sum_i Y_{ji}$). This is actually a simplified representation, because the calculation also takes account of the change in the age-specific incidence rates with absolute time. The expected number of respiratory cancer deaths for the ith exposure group is

$$E(O_j) = \sum_i Y_{ji} a_i (1 + a'D_j^{\,k})/100{,}000$$

$$= E_j(1 + a'D_j^{\,k}) \tag{4}$$

under the relative risk model, and is

$$E(O_j) = \sum_i Y_{ji}(a_i + 100{,}000a'D_j^{\,k})/100{,}000$$

$$= E_j + a'Y_j D_j^{\,k} \tag{5}$$

under the absolute risk model. Consequently, with either model, $E(O_j)$ can be expressed in terms of quantities typically available from the published epidemiologic studies. Note that person-years of observation are not required if the multiplicative model is used.

Making the reasonable assumption that O_j has a Poisson distribution, the parameters a' and k can be estimated from the above equations using the method of maximum likelihood. Once these parameters are estimated, the age-specific mortality rates for respiratory cancer can be estimated for any desired exposure pattern.

To estimate the corresponding additional lifetime probability of respiratory cancer mortality, let $b_1, \ldots, b_{18}$ be the mortality rates, in the absence of exposure, for all cases per year per 100,000 persons for the age intervals 0-4, 5-9,..., 80-84, and 85+, respectively; let $a_1, \ldots, a_{18}$ represent the corresponding rates for malignant neoplasms of the respiratory system. The probability of survival to the beginning of the ith 5-year age interval is estimated as

$$\prod_{j=1}^{i-1} [1 - 5b_j/100,000]. \tag{6}$$

Given survival to the beginning of age interval i, the probability of dying of respiratory cancer during this 5-year interval is estimated as

$$5a_i/100,000. \tag{7}$$

The probability of dying of respiratory cancer given survival to age 85 is estimated as a_{18}/b_{18}. Therefore, the probability of dying of respiratory cancer in the absence of exposure to arsenic is estimated as

$$P_0 = \sum_{i=1}^{17} [5a_i/100,000) \prod_{j=1}^{i-1} (1 - 5b_j/100,000)] \tag{8}$$

$$+(a_{18}/b_{18}) \prod_{j=1}^{17} (1 - 5_{b_j}/100,000)$$

Here the mortality rates a_i apply to the target population for which risk estimates are desired, and consequently will be different from those in (1)-(5), which applied to the epidemiologic study cohort. If the 1976 U.S. mortality rates (male, female, white, and non-white combined) are used in this expression, then $P_0 = 0.0451$.

To estimate the probability P_{EP} of respiratory cancer mortality when exposed to a particular exposure pattern EP, the formula (8) is again used, but a_i and b_i are replaced by $a_i(D_i)$ and $b_i(D_i)$, where D_i is the exposure measure calculated for the ith age interval from the exposure pattern EP. For example, if the dose measure used in (1) is cumulative dose to the beginning of the ith age interval in μg/m³-years, and the exposure pattern EP is a lifetime exposure to a constant level of 10 μg/m³, then $D_i = (i-1)(5)(10)$, where the 5 accounts for the fact that each age inverval has a width of 5 years. The additional risk of respiratory cancer mortality is estimated as

$$P_{EP} - P_0. \qquad (9)$$

If the exposure pattern EP is constant exposure to μg/m³, then $P_{EP} - P_0$ is called the "unit risk."

This approach can easily be modified to estimate the extra probability of respiratory cancer mortality by a particular age due to any specified exposure pattern. It is also clear that the applications of the approach are not limited to respiratory cancer.

7.3.2.2 Risk Estimates from Epidemiologic Studies--Prospective studies of the relationship between mortality and exposure to airborne arsenic have been conducted for the Anaconda, Montana smelter (Lee and Fraumeni, 1969; Lee-Feldstein, 1983; Higgins et al., 1982; Brown and Chu, 1983 a, b, c); and the Tacoma, Washington smelter (Pinto et al., 1977; Enterline and Marsh, 1982).

The study of Lee-Feldstein (1983) reported on an additional 14 years of follow-up of the cohort of 8047 studied by Lee and Fraumeni (1969), and used essentially the same methods of analysis as the earlier study. Therefore, it will not be necessary to consider the Lee and Fraumeni study in any detail in this report. Higgins et al. (1982) followed for an additional 14 years a sample of 1800 men from the cohort studied by Lee and Fraumeni, but used different exposure classifications and different methods of analyses.

Brown and Chu (1983 a, b, c), in a series of papers, arranged the Anaconda smelter data in such a manner that a mathematical model could be derived from it to account for the effect of the timing of exposure as predicted by the multistage model.

Whereas Pinto et al. (1977) studied a cohort from the Tacoma smelter consisting of 527 men followed only after retirement, the study by Enterline and Marsh (1982) involved 2802 men and included follow-up prior to retirement. Consequently, the Enterline and Marsh study appears to provide a stronger basis for quantitative risk assessment than the Pinto et al. study.

In addition to studies of copper smelter workers, there have also been studies of workers exposed to arsenicals in the production or use of pesticides. Ott et al. (1974) studied the mortality experience of workers exposed to lead arsenate and calcium arsenate.

7.3.2.2.1 The Lee-Feldstein (1983) study. This study included 8047 white males who were employed as smelter workers for 12 months or more before 1957, and whose mortality experience was observed from 1938 through 1977. Altogether, the study involved 192,476 person-years of follow-up and 3550 deaths, including 302 from respiratory cancer (Table 7-21). Expected numbers of cancer deaths were calculated on an age-adjusted basis using the combined mortality experience of the white male population of Idaho, Wyoming, and Montana. As Table 7-21 indicates, malignant neoplasms of the digestive and respiratory tracts had SMRs of 125 and 285, respectively, both of which were significant at the 1 percent level (SMR = [observed/expected][100]).

Workers were categorized both by duration of employment and level of exposure to airborne arsenic in order to determine the effect of these parameters upon mortality. For each year of the study period, workers were assigned to one of five groups on the basis of total years of smelter employment completed (Table 7-22). Work areas in the smelter were divided into heavy, medium, or light exposure areas. Based upon this division, workers were categorized into heavy, medium, or light exposure groups, as determined by their maximum exposure for 12 or more months. The results for respiratory cancer based upon these categorizations are given in Table 7-23.

TABLE 7-21. OBSERVED AND EXPECTED DEATHS DUE TO SELECTED CAUSES, WITH STANDARDIZED MORTALITY RATIOS (SMRs) AMONG SMELTER WORKERS, 1938-77

		Number of deaths		
Cause of Death	List No.[a]	Observed	Expected	SMR[b]
Tuberculosis	001-019	53	27.93	190[c]
Respiratory	001-008	47	25.51	184[c]
Other	010-019	6	2.42	248
Malignant neoplasms	140-199	609	370.74	164[c]
Digestive	150-159	167	133.58	125[c]
Respiratory	160-164[d]	302	105.81	285[c]
Other	140-148, 165-170 177-181, 190-199	140	131.35	107
Vascular lesions of central nervous system	330-334	262	211.56	124[c]
Diseases of heart	400-443	1366	1056.55	129[c]
Influenza and pneumonia	480-483, 490-493	88	76.05	116
Emphysema (1963-77 only)	527	90	34.58	260[c]
Cirrhosis of liver	581	76	36.53	208[c]
Accidents	800-962	288	280.27	103
Motor vehicle	810-825, 830-835	106	115.55	92
Other	800-802, 840-862	182	164.72	110
Suicide and homicide	963-964, 970-979 980-985	83	86.08	98
All other causes	Residual	606[e]	548.50	
Total		3522	2728.79	129[c]

[a] Seventh revision of International Lists of Diseases and Causes of Death.

[b] SMR = (observed/expected) x 100.

[c] Significant at 1% level. Bailar and Ederer (1964)

[d] Among the 302 deaths from respiratory cancer, the site was lung and bronchus (162,163) in 289 cases, larynx (163) in 9, mediastinum (164) in 3 and (160) in 1.

[e] Includes 19 emphysema deaths occurring in the years preceding 1963, for which emphysema death rates are not available from individual states.

Source: Lee-Feldstein (1983).

TABLE 7-22. DESCRIPTION OF LENGTH OF EMPLOYMENT GROUPS, WITH NUMBERS OF SMELTER WORKERS, NUMBERS OF DEATHS, PERSON-YEARS AT RISK, AND DURATION OF SMELTER EMPLOYMENT (BASED ON TOTAL WORK EXPERIENCE THROUGH SEPT. 30, 1977)

Length of employment group[a]	Number of persons[b]	Number of deaths	Number of person-years of follow-up[c]
1 (25 or more years)	1899	1169	27,053
2 (15 to 24 years)	1138	586	26,556
3 (10 to 14 years)	678	328	19,734
4 (5 to 9 years)	1082	433	30,854
5 (1 to 4 years)	3248	1006	88,279
TOTAL	8045	3522	192,476

[a] Employees in all cohorts were living on Jan. 1, 1938.

[b] Group assignment of each person here was based on his status at the termination of employment or on September 30, 1977 (whichever date was earlier).

[c] Represents cumulative follow-up experience over the study period, 1938-77, with a total of 67,569 person-years of follow-up in the period 1964-77. Individuals were initially counted at risk upon completing 1 year of employment or on Jan. 1, 1938, if employed at least a full year before that date. In each calendar year of the study period, employees were counted in the group reflecting their cumulative work experience to date.

Source: Lee-Feldstein (1983).

Exposure to airborne arsenic was estimated from 702 samples collected at 56 sampling stations during the years 1943-1958 (Morris, 1975). Morris estimated that airborne levels averaged 11.27, 0.58, and 0.27 mg/m^3 in the heavy, medium, and light exposure areas, respectively. Respirators were used with varying degrees of faithfulness in the high exposure areas; consequently, average individual exposures in these areas were probably much less than 11.27 mg/m^3. A rough estimate is that use of respirators reduces the exposure levels by a factor of 10 (OSHA, 1978).

TABLE 7-23. OBSERVED AND EXPECTED DEATHS FROM RESPIRATORY CANCER, WITH PERSON-YEARS OF FOLLOW-UP, BY COHORT AND DEGREE OF ARSENIC EXPOSURE

	Maximum Exposure to Arsenic (12 or more months)[a]					
	Heavy		Medium		Light	
Years of Exposure	Obs/Exp[b]	P-Y[c]	Obs/Exp	P-Y	Obs/Exp	P-Y
25 years+	13/2.5	2400	49/7	6837	51/16.3	14573
15-24	9/1.3	2629	13/4.0	6509	16/8.6	12520
Less than 15 years	11/2.4	6520	31/9.3	24594	69/31	78245

[a] The 1562 men who worked less than 12 months in their category of maximum arsenic exposure were not included in this table.

[b] Observed/Expected.

[c] Person-years of follow-up furnished by Dr. Lee-Feldstein (personal communication).

Source: Adapted from Lee-Feldstein (1983).

The Lee-Feldstein (1983) study has a number of features which support its use in making quantitative estimates of respiratory cancer risk from airborne arsenic. It was a large study that involved observations of a considerable number of respiratory cancer deaths. A substantial amount of follow-up was conducted of persons who had been exposed for 15 years or more. Estimates of exposure levels and work histories are available for estimating individual exposures and for determining dose response.

It would have been more appropriate for making quantitative estimates of risk to have categorized workers by their individual cumulative or average exposures, rather than by their maximum exposures for 1 year or more. In developing the quantitative estimates, it will be assumed that a worker's average exposure during work hours was equal to the exposure for the category to which he was assigned. However, because these assignments were based upon maximum exposures for at least a 12-month period, this approach tends to overestimate exposures, and consequently, to underestimate the carcinogenic potency of arsenic.

Because smoking is also an important risk factor for respiratory cancer, it would have been very useful to have smoking histories for the workers. Higgins et al. (1982) collected some limited smoking data for this cohort. Higgins and co-workers suggest that the smelter workers smoked somewhat more than the average U.S. white male population, but the difference was not enough to have a major effect upon the outcome of the study.

Development of risk estimates: The data from the Lee-Feldstein (1983) study used in the risk assessment are listed in Table 7-24. The relative risk (observed/expected) from this table are graphed in Figure 7-2, and the absolute risks ([observed-expected/person-years) in Figure 7-3. It is clear from these graphs that the risk for the high-exposure group exposed for greater than 25 years is not commensurate with the risks for the other groups. Because of this, and also because the exposures in the high exposure groups are much more uncertain than those of the other groups, it was decided to estimate risk using only the low and medium exposure groups. Results of applying chi-square goodness-of-fit tests of the relative and absolute risk models with $k = 1$ and $k = 2$ are recorded in Table 7-25. The maximum likelihood estimates of the carcinogenic potency parameter a' are also listed in this table. The maximum likelihood fits of these models are graphed in Figures 7-2 and 7-3. All of the fits are poor (p less than 0.0001) with the exception of that for the absolute-risk model; this latter fit is marginally acceptable ($p = 0.025$).

The unit risk (additional risk of respiratory cancer death from lifetime exposure to 1 $\mu g/m^3$ airborne arsenic) obtained from the absolute-risk model with $k = 1$ is also listed in Table 7-25. This risk was estimated by applying (2), (8), and (9) with D_i based upon a constant exposure of 1 $\mu g/m^3$.

TABLE 7-24. DOSE-RESPONSE DATA FROM LEE-FELDSTEIN (1983) USED FOR RISK ASSESSMENT

Cohort	Maximum Exposure to Arsenic	Cumulative Exposure[a] ($\mu g/m^3$-years)	Person-Years of Observation[b]	Observed Deaths	Expected Deaths
1 (25 + years (of exposure)	Heavy	36064	2400	13	2.5
	Medium	18560	6837	49	7.0
	Light	9280	14573	51	16.3
2 (15-25 years of exposure)	Heavy	22250	2629	9	1.3
	Medium	11600	6509	13	4.0
	Light	5800	12520	16	8.6
3 (less than 15 years of exposure)	Heavy	5973	6520	11	2.4
	Medium	3074	24594	31	9.3
	Light				

[a] Exposures are in $\mu g/m^3$-years estimated as (air concentration)(duration). For light, medium, and heavy exposures, air concentration was estimated as 290, 580, and 1127 $\mu g/m^3$, respectively (OSHA, 1978). Duration was estimated as follows (cf. Table 7-22):

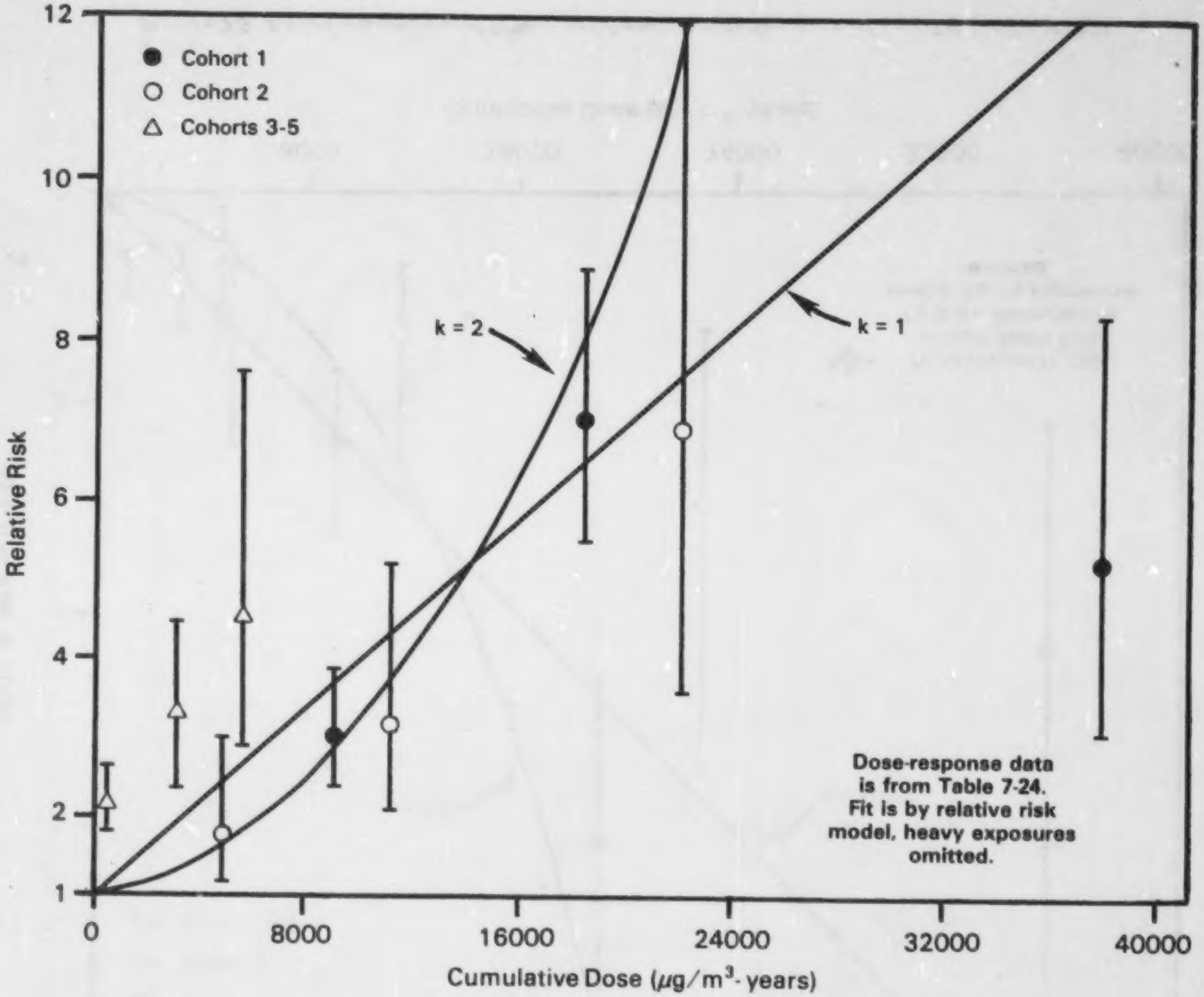
Cohort 1: Persons in this cohort had at least 25 years' exposure. If all had worked continuously throughout follow-up, average duration would have been 25 + 27053/1899 = 39 years. Therefore the midpoint (39 + 25)/2 = 32 years was used.

Cohort 2: The midpoint of the employed interval, i.e., (15 + 25)/2 = 20 years was used.

Cohort 3: A weighted average of the midpoints of the employment intervals, i.e.,

$$\frac{(3)\ (88279) + (7.5)(30854) + (12.5)(19734)}{88279 + 30854 + 19734} = 5.3 \text{ years was used.}$$

[b] Furnished by Dr. Lee-Feldstein.



Dose-response data
is from Table 7-24.
Fit is by relative risk
model, heavy exposures
omitted.

Figure 7-2. Relative risks and 90% confidence limits for data of Lee-Feldstein (1983).

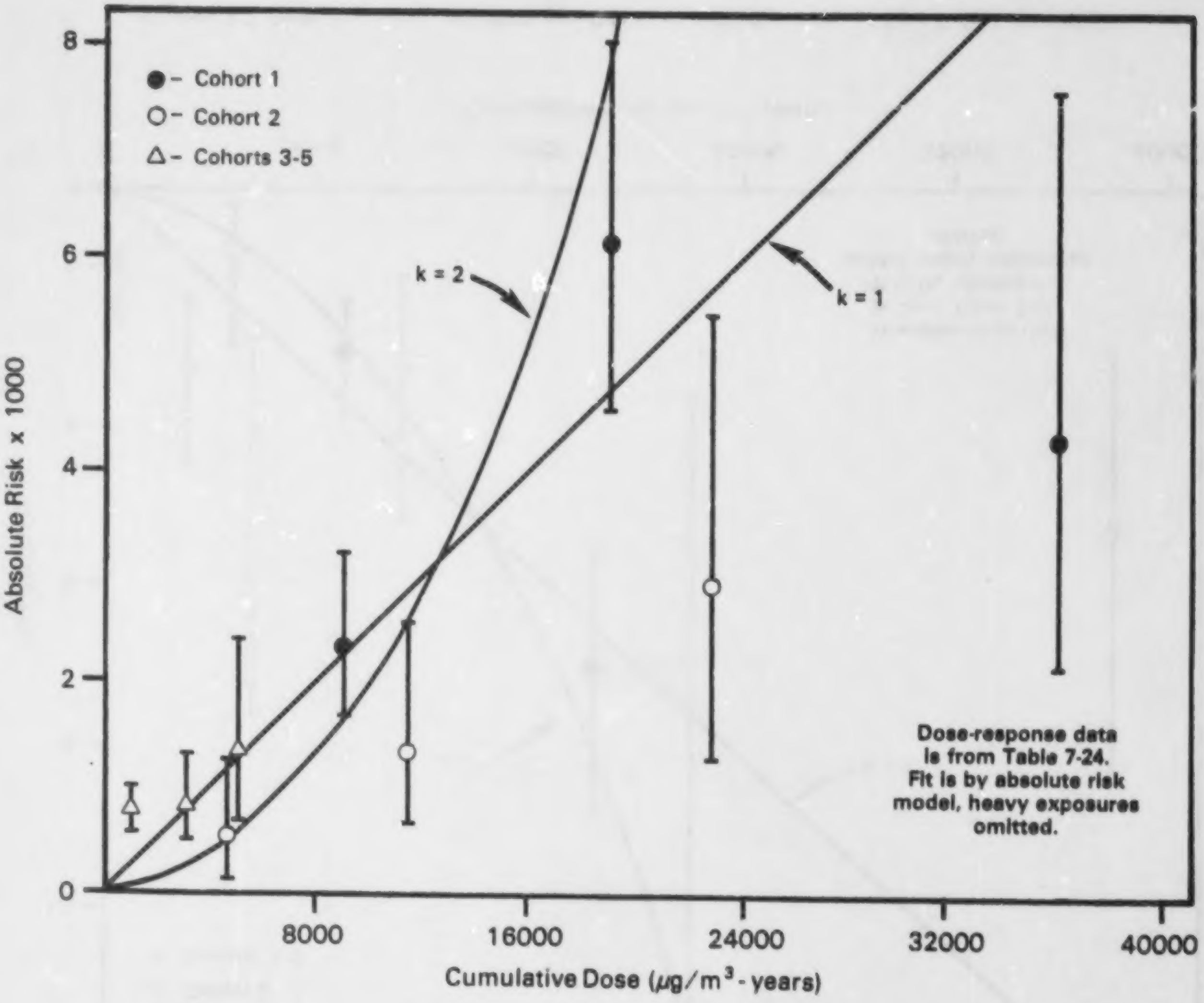


Figure 7-3. Absolute risks and 90% confidence limits for data of Lee-Feldstein (1983).

TABLE 7-25. SUMMARY OF QUANTITATIVE RISK ANALYSES

Exposed Population	Study and Data Source	Model	Results of Goodness-of-Fit Test				
			k	Carcinogenic Potency (a')[a]	x^2 (d.f.)	p-value	"unit" risk[b]
Anaconda smelter workers	Lee-Feldstein Table 7-24 (heavy exposure omitted)	absolute risk	1	2.48(-7)	12.7(5)	0.025	2.80(-3)
			2	2.09(-11)	60(5)	0.00001	lack of fit[d]
		relative risk	1	3.00(-4)	28(5)	0.00004	lack of fit
			2	2.09(-8)	79(5)	0.00001	lack of fit
	Higgins et al. Table 7-26	absolute risk	1	2.36(-7)	1.2(3)	0.75	4.90(-3)
			2	1.74(-11)	10.3(3)	0.017	1.05(-4)
		relative risk	1	3.17(-4)	2.57(3)	0.46	4.03(-3)
			2	2.18(-8)	15.4(3)	0.0015	lack of fit
	Brown & Chu Tables 7-27 and 7-29	absolute risk	1	9.45(-16)	7.01(7)	0.41	1.25(-3)
ASARCO smelter workers	Enterline & Marsh Table 7-33 (zero lag)	absolute risk	1	6.04(-7)[c]	5.5(4)	0.24	6.81(-3)
			2	1.43(-10)	20.7(4)	0.0003	lack of fit
		relative risk	1	4.6(-4)	11.6(4)	0.02	5.44(-3)
			2	1.01(-7)	23.4(4)	0.00011	lack of fit
	Enterline & Marsh Table 7-33 (10-year lag)	absolute risk	1	8.85(-7)	7.0(4)	0.14	7.60(-3)
			2	2.01(-10)	26.1(4)	0.00003	lack of fit
		relative risk	1	5.13(-4)	14.5(4)	0.006	lack of fit
			2	1.06(-4)	28.9(4)	0.00002	lack of fit
Dow pesticide manufacture workers	Ott et al. Table 7-34 (high exposure omitted)	relative risk	1	9.2(-4)	5.0(7)	0.66	1.36(-2)
			2	1.58(-7)	9.4(7)	0.23	7.68(-4)

---

[a] Potencies are in different units, depending upon model used (see text), and consequently are not comparable.

[b] Additional lifetime risk of respiratory cancer mortality from lifetime environmental exposure to 1 μg/m³ arsenic.

[c] 6.04(E-7) means 6.04×10^{-7}.

[d] p-value of chi-square goodness-of-fit test is less than 0.01.

Specifically,

$$D_i = 4.56[(i-1) + 2.5] \quad (10)$$

was used, which represents the cumulative exposure in μg/m³ resulting from a constant exposure to μg/m³ from birth to the midpoint of the ith 5-year age interval. The factor 4.56 is needed to account for the fact that the workers in the occupational study used to estimate the carcinogenic potency of arsenic were only exposed during work hours. Assuming that workers were exposed for an average of 8 hours per day, 240 days per year, an environmental exposure to 1 μg/m³ for 1 year is equivalent to an occupational exposure to

$$(1\ \mu g/m^3)(24\ \text{hours}/8\ \text{hours})(365\ \text{days}/240\ \text{days}) = 4.56\ \mu g/m^3 \quad (11)$$

for 1 year.

7.3.2.2.2 The Higgins et al. (1982) study. Higgins et al. conducted additional independent follow-up through 1977 of a sample of 1800 men from the Anaconda smelter cohort studied by Lee and Fraumeni (1969). The sample included all of the men classified in the heavy exposure category by Lee and Fraumeni (1969), as well as a random sample of 20 percent of the remaining cohort. There were 80 deaths from respiratory cancer in the sample. Expected numbers were based upon the mortality rates of Montana white males, except for "all respiratory diseases"; U.S. white males were the referenced population for the latter category.

Higgins et al. also reviewed the industrial hygiene data and calculated average concentrations for the period 1943-1965 for 18 departments. No measurements were available for 17 departments, and average concentrations were estimated for these. These estimates were coupled with work histories updated through 1978 to obtain exposure measures for each individual in the study. Three types of individual exposure measures were considered: ceiling, time-weighted average (TWA), and cumulative. Ceiling exposures were estimates, in $\mu g/m^3$, of the highest exposure a man experienced for 30 days or more. TWA exposures were estimates, in units of $\mu g/m^3$, of the time-weighted average exposures during the period of employment. Cumulative exposures were estimates of total exposure in units of $\mu g/m^3$-years.

Higgins et al. investigated 5 combinations of follow-up and time period during which exposure was assessed: I. Exposure was assessed up to the date a worker entered the study and follow-up was from entry into the study through 1978; II. Exposure was assessed up to 1964 and follow-up was also through this date; III. Exposure was assessed through 1964 and follow-up was through 1978; IV. Exposure was assessed through 1964 and follow-up was from 1964 through 1978; V. Exposure was assessed through 1978 and follow-up was also through 1978. These different methods were considered principally because of the perceived difficulties of overlapping exposure and follow-up periods. Thus, with methods I and IV, exposure and follow-up periods were disjoint, whereas with methods II and V they coincided.

The analyses based upon ceiling exposures are not considered suitable for quantitative risk assessment because it seems extremely unlikely that respiratory cancer risk would be a function of peak exposures for any 30-day period, regardless of the other exposures that might have been experienced. This is also the case with the TWA analyses, because the exposures were averaged only over the period of employment, without regard to the duration of employment. If either of these dose measures were appropriate, it would mean, for example, that exposure to 500 $\mu g/m^3$ for 1 month would produce the same risk as exposure to 500 $\mu g/m^3$ for 30 years--which seems highly unlikely.

Thus, the analyses of Higgins et al. which appear to be most appropriate for developing quantitative estimates are those based upon cumulative exposure. This particular type of analysis was applied only to method V and applied only in Higgins (1982). The results of this analysis for lung cancer are listed in Table 7-26.

TABLE 7-26. RESPIRATORY CANCER MORTALITY 1938-1978 FROM CUMULATIVE EXPOSURE TO ARSENIC FOR 1800 MEN WORKING AT THE ANACONDA COPPER SMELTER[a]

Cumulative Exposure $\mu g/m^3$-years	Person-Years of Observation	Observed Deaths	Expected Deaths
0-500 (250)[b]	13845.9	4	5.8
500-2000 (1250)	10713.0	9	5.7
2000-12000 (7000)	11117.8	27**	6.8
≥ 12000 (16000)	9015.5	40**	7.3

[a] From Higgins (1982), Table 6.

[b] Numbers in parenthesis indicate assumed average exposures.

** Significant at 0.01 level.

Information on the smoking habits of 80.6 percent of the 1800 men was obtained from questionnaires administered directely to those still living and to close friends or relatives of those who were deceased. Sixteen percent of the smelter workers were "non-smokers" compared with 24-36 percent of U.S. males from 1955 through 1978. Thus, it appears that the smelter workers smoked somewhat more than the average U.S. male. However, no confounding was detected between arsenic exposure and smoking; 15.1 percent of those in the "heavy" exposure group were non-smokers, versus 16.3 percent in the other exposure groups. Significant increases in respiratory cancer were observed even among non-smokers exposed to high levels of arsenic.

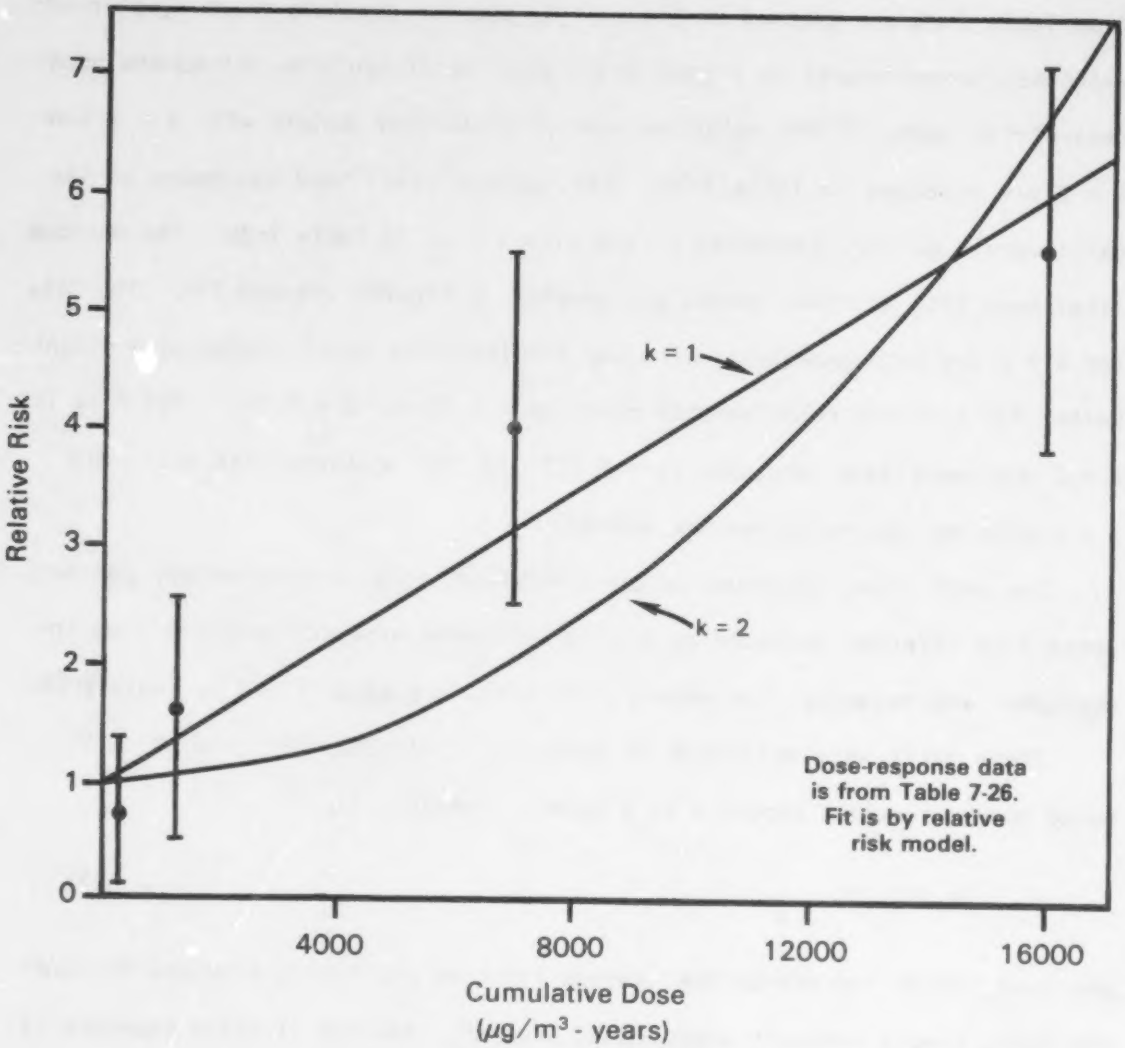
Development of risk estimates: The relative risks (observed/expected) from Table 7-26 are graphed in Figure 7-4, and the absolute risks ([observed-expected]/person-years) in Figure 7-5. Results of applying chi-square goodness-of-fit tests of the relative- and absolute-risk models with $k = 1$ and $k = 2$ are recorded in Table 7-25. The maximum likelihood estimates of the carcinogenic potency parameter a' are also listed in Table 7-25. The maximum likelihood fits of those models are graphed in Figures 7-4 and 7-5. The fits for $k = 1$ are both excellent, with the absolute-risk model providing a slightly better fit than the relative-risk model ($p = 0.75$ vs. $p = 0.46$). The fits for $k = 2$ are much less adequate ($p = 0.017$ for the absolute-risk model and $p = 0.0015$ for the relative-risk model).

The unit risks (defined as the additional risk of respiratory cancer death from lifetime exposure to 1 $\mu g/m^3$ airborne arsenic) obtained from the absolute- and relative-risk models with $k = 1$ are also listed in Table 7-25.

These risks were estimated by applying (1) or (2), (8), and (9) with D_i based upon a constant exposure of 1 $\mu g/m^3$. Specifically,

$$D_i = (4.56)(72) \qquad (12)$$

was used, which represents the average lifetime cumulative exposure in $\mu g/m^3$ resulting from a constant exposure to 1 $\mu g/m^3$. Average lifetime exposure is used because it seems most commensurate with the treatment of exposure by Higgins et al.; in their analysis all of the person-years attributable to a single worker were placed into a single exposure category based upon total lifetime exposure, and consequently person-years of observation were placed into exposure categories according to exposures which had not yet occurred. It



Dose-response data is from Table 7-26. Fit is by relative risk model.

Figure 7-4. Relative risks and 90% confidence limits for data of Higgins (1982).

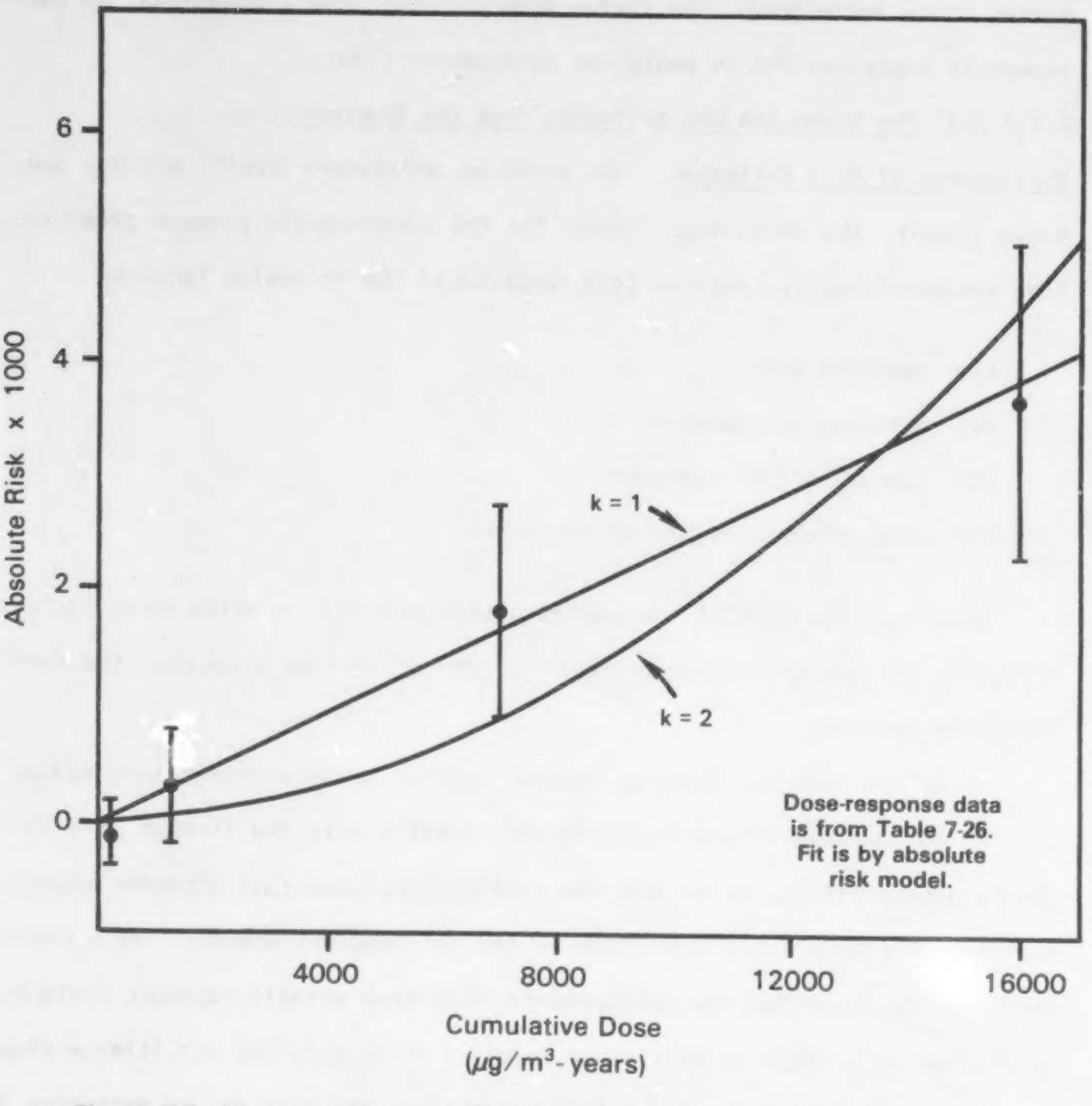


Figure 7-5. Absolute risks and 90% confidence limits for data of Higgins (1982).

would have been more appropriate for purposes of quantitative risk assessment had exposures been related to each 5-year age interval (as was done in the analysis of Enterline and Marsh, 1982) rather than to the total observation period of an individual. The factor 4.56 converts from occupational to environmental exposures and is explained at equation (11).

7.3.2.2.3 The Brown and Chu estimates from the Anaconda data.

Development of Risk Estimates. As noted by Whittemore (1977) and Day and Brown (1980), the multistage theory for the carcinogenic process predicts that the carcinogenic response is a function of the following factors:

(1) exposure rate
(2) duration of exposure
(3) age at initial exposure
(4) time since cessation of exposure.

Brown and Chu (1983a) discuss in detail the ways in which these factors influence the age-specific carcinogenic rate at various stages of the carcinogenic process.

Using the updated Anaconda copper smelter workers cohort originally studied by Lee and Fraumeni (1969) and recently extended through 1977 by Lee-Feldstein (1983), Brown and Chu (1983b) concluded that airborne arsenic most probably acted on a late stage of the carcinogenic process. As a result, they hypothesized that the carcinogenic risk from arsenic exposure could be quantified by assuming a multistage model in which only the penultimate stage is affected by exposure. Under this assumption, the risk may be expressed in the form

$$r(d, t_o) = C[(d + t_o)^{k-1} - t_o^{\ k-1}] \qquad (13)$$

where d is the duration of exposure, t_0 is the age at initial exposure, and C and k are unknown parameters. The parameter C depends upon the exposure rate, and the parameter k upon the time effect of exposure.

Brown and Chu (1983b) noted a deviation from this model on the part of workers who left employment at the copper smelter before the age of 55. As a result, the mortality experience of that group after leaving employment was not included in the analysis.

In order to estimate the unknown parameters C and k, the basic mortality data were arranged in the three-way table reproduced as Table 7-27. The three classifications used in this table are as follows:

(1) Level of exposure, corresponding to Lee and Fraumeni (1969)--classified into heavy, medium, and light exposure groups;

(2) Duration of employment, classified into the following five subgroups; 0-9, 10-19, 20-29, 30-39, and 40+ years;

(3) Age at initial employment, classified into the following five subgroups; 20, 20-29, 30-39, 40-49, and 50+ years.

For each of the 3 x 5 x 5 = 75 cells in the table, the following three variables were given:

Obs = observed number of respiratory cancer deaths;

Exp = expected number of respiratory cancer deaths (based upon the U.S. white male age-specific calendar-time-specific respiratory cancer mortality rates); and

Pyr = person-years of observation

A single individual could supply information for more than one cell as his duration of employment increased over the follow-up period. The person-year weighted average duration of employment, and age at initial employment, were calculated for each cell.

Assuming that age at initial exposure is equivalent to age at initial employment, and that duration of employment and exposure are equivalent, Brown and Chu (1983c) fitted equation 13 to the data in Table 7-27. They used the maximum likelihood method, assuming a binomial distribution where Obs is the number of positive responses, Pyr is the sample size, and the rate of response is $p = \text{Exp}/\text{Pyr} + r(\bar{d},\bar{t}_o)$, where $\bar{d},\bar{t}_o$ are the averages for each cell. Using this approach, the value for k is estimated to be 6.8, and c = .603, 1.42, 1.74×10^{-13} for the light, medium, and heavy exposure categories, respectively.

Brown and Chu (1983c) did not attempt to give an exposure rate estimate to the heavy, medium, and light exposure groups of Lee and Fraumeni, (1969). One reason for this was that "heavy" and "medium" were defined as "having worked at least one year in a heavy or medium exposure area." The "total time worked" was not necessarily an indication of the total time worked in a heavy or medium exposure area. As a result, the use of the exposure rate in the areas defined as medium or heavy would tend to overestimate the true average exposure over an individual's working history. This bias did not exist for those in the light exposure group, since almost all of their working time was spent in light exposure areas. In addition, these low environmental exposures are of greater utility in estimating risks.

As a result of these factors, only the light exposure group was used to obtain a dose-response model. In this group, Brown and Chu (1983c) estimated that the respiratory cancer rate for an individual first exposed at age t_o for a duration of d years would be

$$r(d,t_o) = .603 \times 10^{-13} \left[(d + t_o)^{5.8} - t_o^{5.8}\right]. \qquad (14)$$

Only limited information exists concerning the time-weighted exposure of workers in the light exposure areas. Arsenic concentrations in several light

TABLE 7-27. OBSERVED AND EXPECTED LUNG CANCER DEATHS AND PERSON-YEARS BY LEVEL OF EXPOSURE, DURATION OF EMPLOYMENT, AND AGE AT INITIAL EMPLOYMENT

Age at Initial Employment		Duration of Employment (years)				
		0-9	10-19	20-29	30-39	40+
High Exposure Level Group						
<20	Obs	0	0	0	3	0
	Exp	0.001	0.009	0.065	0.249	0.193
	Pyr	206	408	588	499	172
20-29	Obs	0	0	2	0	2
	Exp	0.008	0.051	0.164	0.277	0.082
	Pyr	624	637	495	308	64.4
30-39	Obs	0	0	3	0	0
	Exp	0.030	0.077	0.106	0.053	0.001
	Pyr	398	207	155	59.1	0.86
40-49	Obs	0	0	0	0	0
	Exp	0.083	0.054	0.034	0.007	0.0
	Pyr	210	80.0	49.1	6.88	0.0
50+	Obs	0	0	0	0	0
	Exp	0.066	0.027	0.0	0.0	0.0
	Pyr	78.0	23.2	0.0	0.0	0.0
Medium Exposure Level Group						
<20	Obs	0	0	1	4	1
	Exp	0.010	0.039	0.171	0.591	0.597
	Pyr	1801	1763	1500	1206	579
20-29	Obs	0	0	2	4	7
	Exp	0.035	0.118	0.331	0.717	0.514
	Pyr	2636	1622	1099	951	654
30-39	Obs	0	0	1	3	0
	Exp	0.167	0.473	0.329	0.161	0.045
	Pyr	1939	1137	438	194	68.2
40-49	Obs	0	0	1	3	0
	Exp	0.167	0.414	0.098	0.010	0.0
	Pyr	1190	448	98.9	12.1	0.0
50+	Obs	1	0	0	0	0
	Exp	0.262	0.076	0.011	0.0	0.0
	Pyr	295	71.2	14.5	0.0	0.0

TABLE 7-27. (continued)

Age at Initial Employment		Duration of Employment (years)				
		0-9	10-19	20-29	30-39	40+
Low Exposure Level Group						
<20	Obs	0	0	1	1	3
	Exp	0.056	0.117	0.478	1.59	1.57
	Pyr	8524	5249	4038	3175	1376
20-29	Obs	0	0	2	5	6
	Exp	0.115	0.334	0.892	1.74	0.796
	Pyr	9951	4724	2965	2117	834
30-39	Obs	0	3	1	0	1
	Exp	0.390	0.802	0.937	0.662	0.062
	Pyr	5218	2218	1364	715	74.6
40-49	Obs	2	1	1	1	0
	Exp	1.29	1.18	0.344	0.035	0.001
	Pyr	3703	1319	386	52.7	2.00
50+	Obs	3	2	0	0	0
	Exp	1.62	0.385	0.041	0.0	0.0
	Pyr	1945	371	65.4	0.0	0.0

Source: Brown and Chu (1983a).

exposure areas, as given in a NIOSH criteria document (1975), are shown in Table 7-28.

In the absence of information to the contrary, it is assumed that the person-hours spent in each area are equal. Thus an estimate of the time-weighted average for workers in the light exposure category is

$$1/3 \times .7 + 1/3 \times .17 + 1/3 \times .004 = .291 \text{ mg As/m}^3.$$

Under the linear assumption, equation 14 may be expressed in terms of mg As/m^3 working exposure by dividing by .291, which gives the result

$$r(d,t_o) = 2.07 \times 10^{-13} [(d + t_o)^{5.8} - t_o^{5.8}]. \qquad (15)$$

TABLE 7-28. ARSENIC EXPOSURES: 1965 SMELTER SURVEY ATMOSPHERIC ARSENIC CONCENTRATIONS (mg/As/m^3)

"Heavy exposure area" as classified by Lee and Fraumeni				
Arsenic Roaster Area			Mean:	1.47
	0.10	0.20	Median:	0.185
	0.10	0.22		
	0.10	0.25		
	0.10	0.35		
	0.10	1.18		
	0.10	5.00		
	0.17	12.66		
"Medium exposure areas" as classified by Lee and Fraumeni				
Reverberatory Area			Mean:	1.56
	0.03	0.93	Median:	0.88
	0.22	1.00		
	0.23	1.27		
	0.36	1.60		
	0.56	1.66		
	0.63	1.84		
	0.66	1.94		
	0.76	2.06		
	0.78	2.76		
	0.78	3.40		
	0.80	4.14		
	0.83	8.20		
Treater Building and Arsenic Loading			Mean:	1.50
	0.10	0.48	Median:	0.295
	0.10	0.62		
	0.10	3.26		
	0.11	7.20		
"Light exposure areas" as classified by Lee and Fraumeni				
Copper Concentrate Transfer System			Mean:	0.70
	0.25		Median:	0.65
	0.65			
	1.20			
Samples from Flue Station			Mean:	0.17
	0.10		Median:	0.17
	0.24			
Reactor Building			Mean:	0.004
	0.001	0.003	Median:	0.002
	0.002	0.009		
	0.002	0.010		
	0.002			

Source: Table X-3, NIOSH Criteria Document (1975).

In this case, exposure is expressed in mg As/m^3 per 8-hr working day. To change the relationship so that it expresses the risk due to a lifetime of continuous exposure to 1 ug As/m^3, we assume 240 days worked per year, one mg As/m^3 on the job gives the same cumulative exposure as $10^3 \times 1/3 \times \frac{240}{365} = 219$ μg As/m^3 continuous exposure.

The age-specific rate due to a continuous 1 μg As/m^3 exposure is obtained by substituting $t_o = 0$ and $d = t =$ age into equation (15) and dividing by 219 to arrive at the correct number of exposure units. This gives the result

$$r(t) = 9.45 \times 10^{-16} \, t^{5.8}. \qquad (16)$$

The unit risk is approximately equivalent to the risk of induced respiratory cancer in the median life span. Based upon 1976 U.S. vital statistics, the median life span is 76.2 years, so that the unit risk is expressed approximately as

$$P \simeq \int_0^{76.2} 9.45 \times 10^{-16} \, t^{5.8} = \frac{9.45}{6.8} (76.2)^{6.8} \times 10^{-16} = 8.71 \times 10^{-4}. \qquad (17)$$

An additional approximation consistent with the previous unit calculations is obtained by assuming that

$$r(t) = 9.45 \times 10^{-16} \left[\frac{t_j + t_j-1}{2}\right]^{5.8} \quad t_{j-1} < t < t_j \qquad (18)$$

where t_j are the ages at the interval boundaries given in U.S. vital statistics records. This assumes that the age-specific death rate due to the exposure is constant throughout the interval and equal to the true value at the midpoint of the interval. Under this approximation, using 1976 vital statistics, the unit risk is estimated to be $P \simeq 1.25 \times 10^{-3}$.

Evaluation of Goodness-of-Fit. It is desirable to assess whether the data for the low-exposure group is consistent with the model utilized to estimate the unit risk. However, two factors tend to create a situation that would decrease this goodness-of-fit and bias the results. First, the exact values for each cell of $\bar{d}$, $\bar{t}_o$ are presently not available. Second, the value of k was determined on the basis of all three exposure groups, and does not give as good a fit as would be obtained using the low-exposure group alone.

Brown states (personal communication, 1983) that the exact values of $\bar{d},\bar{t}_o$ are very close to the midpoint of the interval, and that the values of k appear to be statistically consistent between exposure groups. Thus, distortions of the data because of the use of midpoint values and average k, although inevitable, are not appreciable.

The expected number of cases in each cell are calculated using the relationship

$$E = \text{Exp} + \text{Pyr} \times .603 \times 10^{-13} \left[(d + t_o)^{5.8} - t_o^{5.8}\right], \qquad (19)$$

in which Exp and Pyr are taken from Table 7-27 and d,t_o are the midpoints of the intervals. These results are shown in Table 7-29. A standard chi-square goodness-of-fit test is then run, resulting in a chi-square value of X^2_{21} = 13.85 with 23.2 = 21 degrees of freedom and an associated p-value of .88. Unfortunately, due to the low expected number of cases in many of the cells, the X^2 approximation is of questionable validity for this situation.

To obtain a more stable approximation, cells with low frequency that are as close as possible to each other are usually combined. It is important to have some criteria for combining the data that do not depend upon inspection of the data itself. Two methods of combining the data are used here. The first is across columns (duration exposed), so that the maximum number of

TABLE 7-29. OBSERVED AND EXPECTED NUMBER OF RESPIRATORY CANCER DEATHS FOR EACH CELL IN THE LOW-EXPOSURE GROUP OF TABLE 7-27

	d				
t_o	5	15	25	35	45
18	0 .088	0 .314	1 1.20	1 3.504	3 3.836
25	0 .258	0 .858	2 2.145	5 4.358	6 3.321
35	0 .723	3 1.641	1 2.556	0 2.791	1 .497
45	2 2.023	1 2.508	1 1.428	1 .371	0 .026
55	3 2.582	2 1.234	0 .418		

$X^2_{21} = 13.85$, p = .88

cells are obtained, with the constraints that combined cells must have at least three expected cases, and that all cells combined are consecutive within a row. The second approach uses the same technique within a column (age first exposed). The results are shown in Tables 7-30 and 7-31 respectively, giving chi-square values of 7.01 and 7.61, with p-values of .41 and .38. Thus the assumed model is shown to be consistent with the observed low-exposure data.

7.3.2.2.4 The Enterline and Marsh (1982) study. This study included all men (2802 in all) employed at the Tacoma, Washington copper smelter for a year or more during 1940-1964. Their mortality experience was observed through 1976. The study involved over 70,000 person-years of observation, and 104 deaths from cancer of the respiratory system were recorded (Table 7-32). Respiratory cancer deaths had an SMR of 189.4, which was significantly increased at the 1 percent level. Expected deaths for Table 7-32 were based upon U.S. white

TABLE 7-30. CELLS FROM TABLE 7-29 COMBINED WITHIN ROWS TO OBTAIN CELLS WITH THREE OR MORE EXPECTED RESPIRATORY CANCER DEATHS

	d				
t_o	5	15	25	35	45
18				2 5.106	3 3.836
25			2 3.261	5 4.358	6 3.321
35			4 4.920		1 3.288
45					5 6.356
55			5 4.234		

$X^2 = 7.01$, $p = .41$

TABLE 7-31. CELLS FROM TABLE 7-29 COMBINED WITHIN COLUMNS TO OBTAIN CELLS WITH 3 OR MORE EXPECTED RESPIRATORY CANCER DEATHS

	d				
t_o	5	15	25	35	45
18				1 3.504	3 3.836
25			3 3.345	5 4.358	
35					
45				1 3.162	7 3.844
55	5 5.674	6 6.555	2 4.402		

$X^2 = 7.61$, $p = .38$

TABLE 7-32. OBSERVED DEATHS AND SMRs FOR 2802 SMELTER WORKERS WHO WORKED A YEAR OR MORE 1940-64, FOLLOWED THROUGH 1976, BY CAUSE OF DEATH

Cause of death (7th revision code)	Observed Deaths	SMR
All causes of death	1061	103.2
Tuberculosis (001-019)	4	27.6**
Malignant neoplasms (140-148)	231	123.6**
Buccal cavity and pharynx (140-148)	7	110.7
Digestive organs & peritoneum (150-159)	65	108.9
Esophagus (150)	3	66.2
Stomach (151)	17	122.1
Large intestine (153)	21	120.4
Rectum (154)	9	122.4
Biliary passages and liver (155-156)	3	64.1
Pancreas (157)	11	106.0
All other digestive organs (residual)	1	71.6
Respiratory system (160-164)	104	189.4**
Larynx (161)	2	67.7
Bronchus, trachea, and lung (162-163)	100	194.9**
All other respiratory system (residual)	2	305.0
Prostate (177)	11	79.0
Testes and other genital (178-179)	1	92.6
Kidney (180)	6	133.3
Bladder and other urinary organs (181)	4	63.0
Malignant melanoma of skin (19)	0	--
Eye (192)	1	492.7
Central nervous system (193)	3	59.8
Thyroid gland (194)	0	--
Bone (196)	2	175.0
Lymphatic & haematopoietic (200-205)	17	93.8
Lymphosarcoma and reticulosarcoma (200)	4	93.2
Hodgkins' disease (201)	2	83.9
Leukemia and aleukemia (204)	6	78.7
Other lymphopoietic tissue (202, 203, 205)	5	130.4
Other malignant neoplasms (residual)	10	82.3
Benign neoplasms (210-239)	2	78.6
Diabetes mellitus (260)	12	84.8
Stroke (333-334)	91	111.4
Heart disease (400-443)	412	92.5
Hypertension without heart disease (444-447)	1	18.8
Nonmalignant respiratory disease (470-527)	60	108.6
Influenza and pneumonia (480-493)	24	92.9
All other respiratory diseases (residual)	36	122.4
Ulcer of stomach and duodenum (540-541)	7	75.5
Cirrhosis of liver (581)	22	101.9
Chronic nephritis (592)	6	87.5
External causes of death (800-998)	81	94.2
Accidents (899-962)	61	100.6
Suicides (963, 970-979)	17	84.8
Other external causes (residual)	3	56.2
Other causes of death (residual)	85	86.1
Unknown causes	47	--

* $p < .05$, ** $p < .01$

Source: Enterline and Marsh (1982).

male mortality rates. The respiratory cancer SMR increases to 198.1 when Washington State mortality rates are applied.

Enterline and Marsh estimated individual exposures to airborne arsenic using individual work histories, urine arsenic measurements, and an estimated correlation between exposure to airborne arsenic and resulting levels of arsenic in urine. Average urine arsenic levels were available by department for the years 1948-52, 1973, 1974, and 1975. Linear interpolations were used to estimate levels between 1952 and 1973. Levels during 1949-1952 were assumed to hold prior to that time. By coupling these data with employee work histories, Enterline and Marsh estimated individual cumulative exposures for various times in units of μg-years/ℓ urinary arsenic.

Pinto et al. (1977) compared airborne concentrations of arsenic with urinary arsenic levels for 24 workers wearing personal air samplers for 5 successive days. A regression analysis of these data showed a highly significant linear correlation between airborne and urinary arsenic ($p < 0.01$). Average airborne arsenic in units of $\mu g/m^3$ was estimated to be 0.304 times average urinary arsenic levels in units of μg/ℓ.

To investigate dose-response, Enterline and Marsh divided the total person-years of observation into 5 groups by cumulative arsenic exposure (0 lag), and also by cumulative arsenic exposure up to 10 years prior to the year of observation (10-year lag). In this type of analysis, as a worker continues to be exposed to arsenic, he or she will contribute person-years to progressively higher exposure categories. The numbers of respiratory cancer deaths and corresponding expected numbers for each of these groups are given in Table 7-33. In this table, urinary arsenic levels provided by Enterline and Marsh have been converted to airborne exposures in $\mu g/m^3$-years, using the factor 0.304 estimated by Pinto et al. (1977). The observed numbers of cancers are all significantly increased at the higher exposure levels.

TABLE 7-33. DATA FROM TABLE 8 OF ENTERLINE AND MARSH (1982) WITH PERSON-YEARS OF OBSERVATION ADDED

Cumulative Exposure[a] μg/m^3-years	Person-Years of Observation[b]	Observed Deaths	Expected Deaths
	0 Lag		
91.8	10902	8	4.0
263	21642	18	11.0
661	14623	21	10.3
1381	13898	26	14.1
4091	9398	31	12.7
	10-Year Lag		
91.8	27802	10	6.4
263	16453	22	12.5
661	11213	26	11.5
1381	9571	22	12.4
4091	5423	24	9.7

[a] Exposures are in μg/m^3-years estimated by the formula (I μg/l-years) (0.304) where I is mean urinary exposure index from Enterline and Marsh (1982) Table 8 and 0.304 is the relation between urinary and airborne arsenic estimated by Pinto et al. (1977).

[b] Furnished by Dr. Enterline (personal communication).

Although the Enterline and Marsh (1982) study is not as large as that of Lee-Feldstein (1983), it does involve a sizable number of respiratory cancer deaths (104). Workers were followed for an extended period--an average of 25 years per individual. Several features of the analysis render it more amenable to quantitative risk estimation than the analysis used by Lee-Feldstein. Enterline and Marsh made estimates of individual exposure histories, whereas Lee-Feldstein did not. The type of dose-response analysis used by Enterline and Marsh is also more suitable for quantitative risk estimation. The exposure estimates based on a 10-year lag probably yield a more realistic dose response than those that do not utilize a lag. Because the latency period for respiratory cancer is generally greater than 10 years (cf. Doll and Peto, 1978), exposure during the last 10 years prior to observation would not be expected to affect respiratory cancer mortality.

By applying urinary arsenic measurements made during the years 1948-52 to earlier years, Enterline and Marsh probably underestimated exposures prior to 1948. This would result in an overestimate of the carcinogenic potency of arsenic. A calculation by Enterline and Marsh of SMRs by both year of hire and cumulative exposure indicates that workers in a given cumulative exposure category tend to have at least roughly comparable SMRs irrespective of the year of hire. This suggests that exposure estimates for earlier years are not greatly in error. However, further investigation of this problem would be useful.

Because smoking is also an important risk factor for respiratory cancer, it would have been helpful if data on smoking habits had been available for analysis. Pinto and Enterline (undated) report on smoking histories obtained in 1975 from 550 active employees at the Tacoma smelter. Of these employees, 59.6 percent were active smokers, compared to 45.4 percent in 1970 for U.S. males aged 21-64. If this excess of smokers holds in general for the smelter workers, then a small fraction of the excess in respiratory cancer could have been due to smoking.

Development of risk estimates: The data from Table 7-33 were used for quantitative risk assessment. The relative risks (observed/expected) from this table are graphed in Figures 7-6 and 7-7, and the absolute risks ([observed- expected]/person-years) in Figures 7-8 and 7-9. Although there is no clear trend of increasing SMRs with increasing exposures, such a trend is present for absolute risk. Results of applying chi-square goodness-of-fit tests of the relative- and absolute-risk models with $k = 1$ and $k = 2$ are recorded in Table 7-25. The maximum likelihood estimates of the carcinogenic

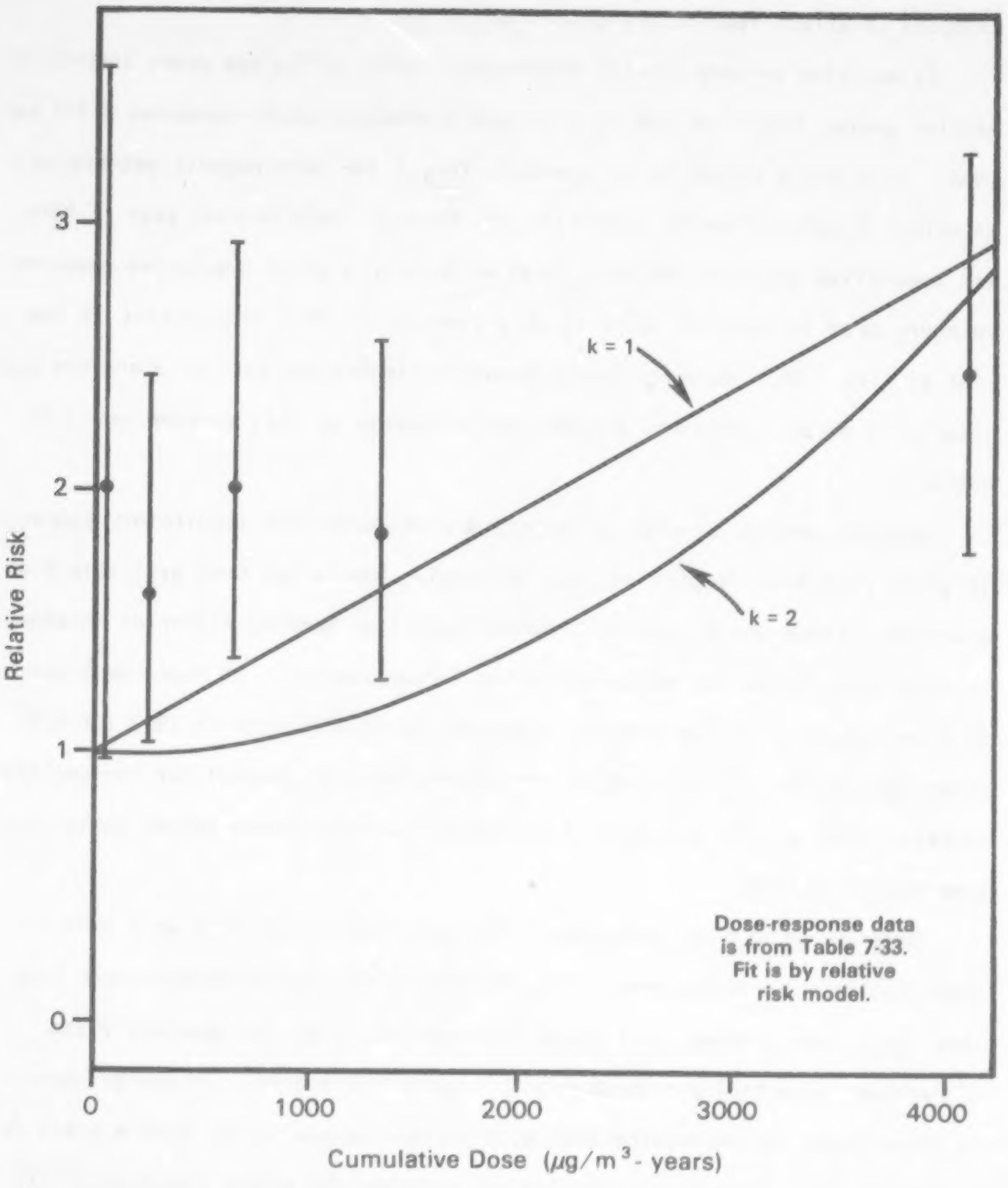


Figure 7-6. Relative risks and 90% confidence limits for zero-lag data of Enterline and Marsh (1982).

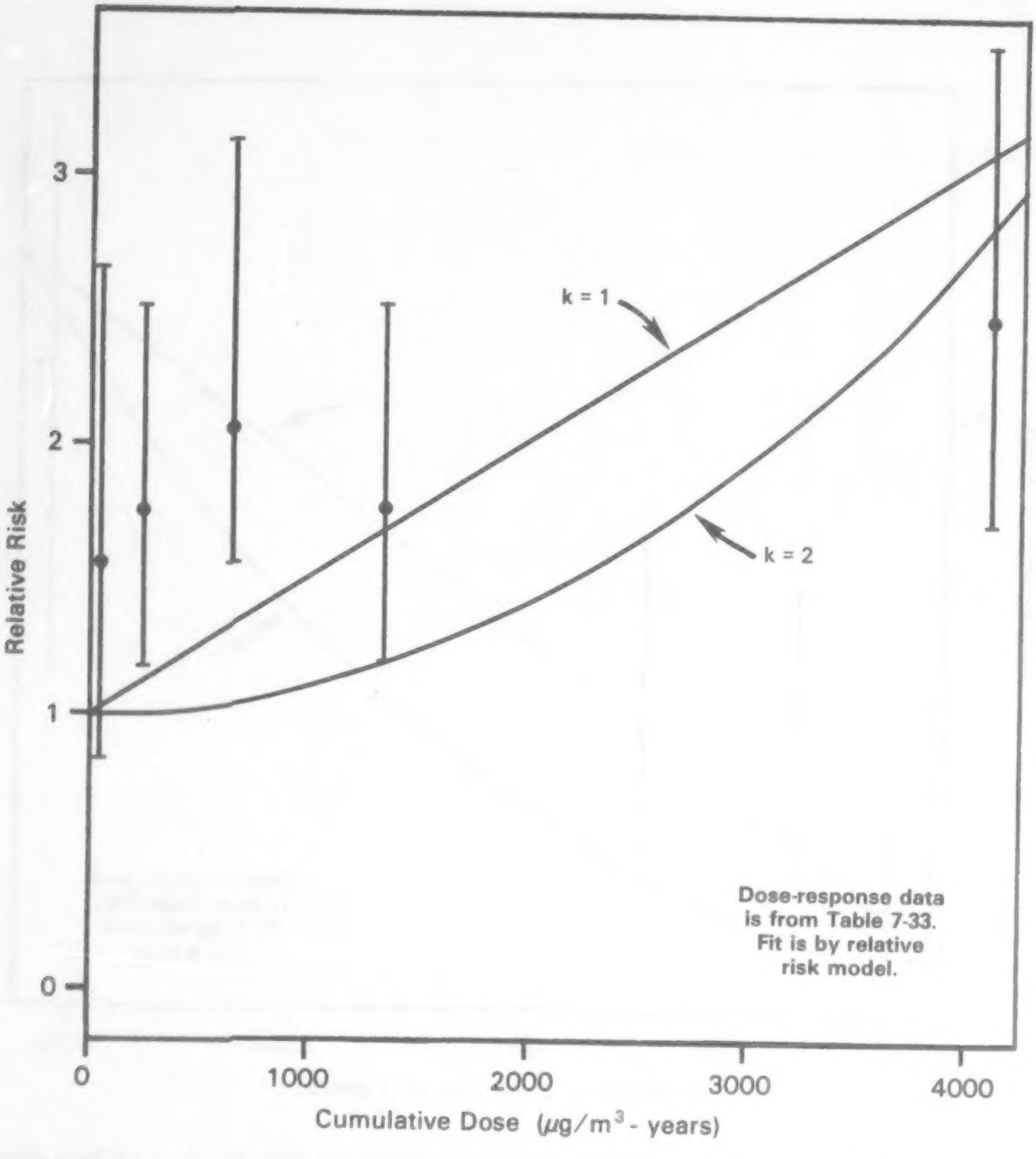


Figure 7-7. Relative risks and 90% confidence limits for 10 year lag data of Enterline and Marsh (1982).

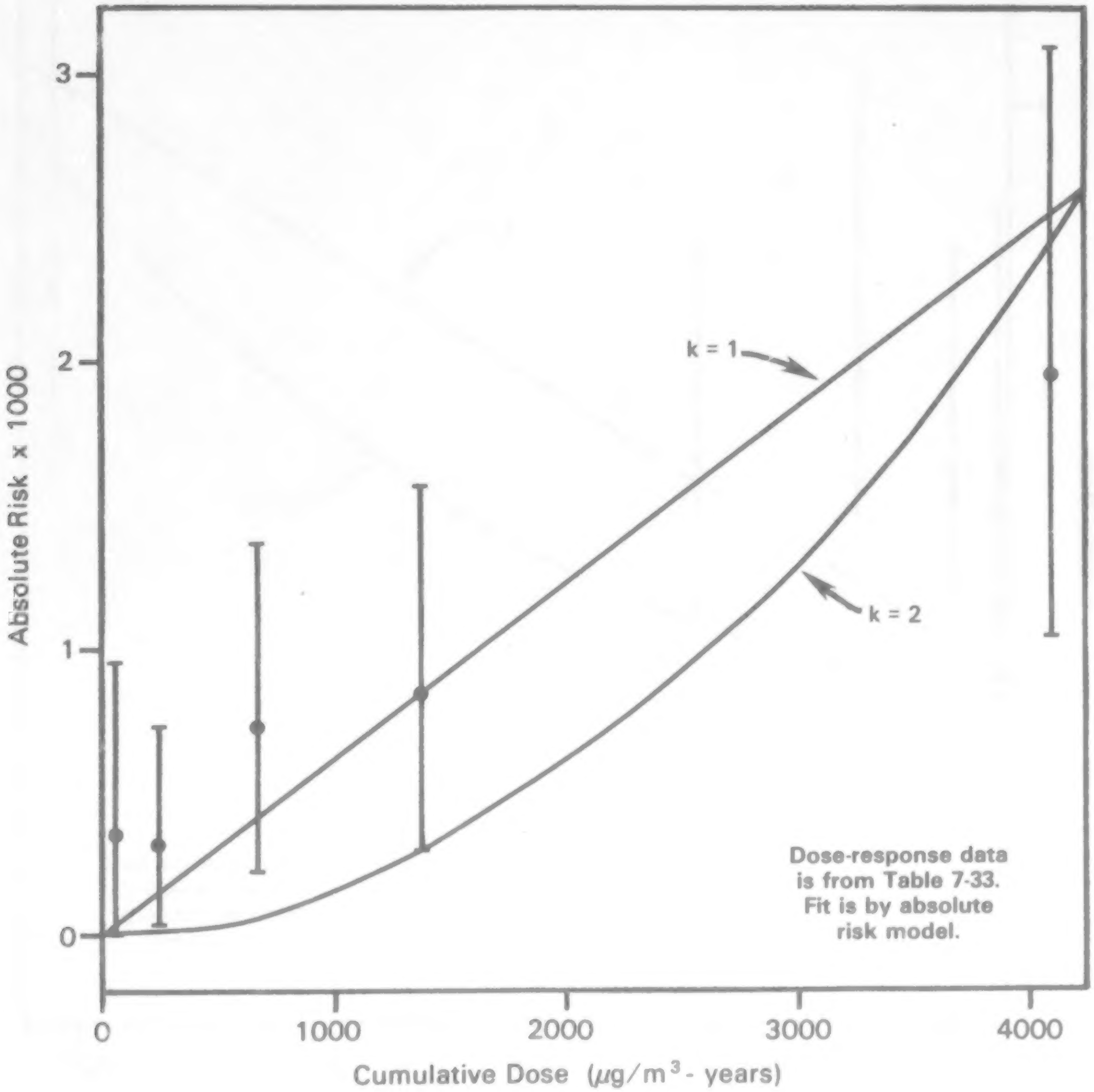
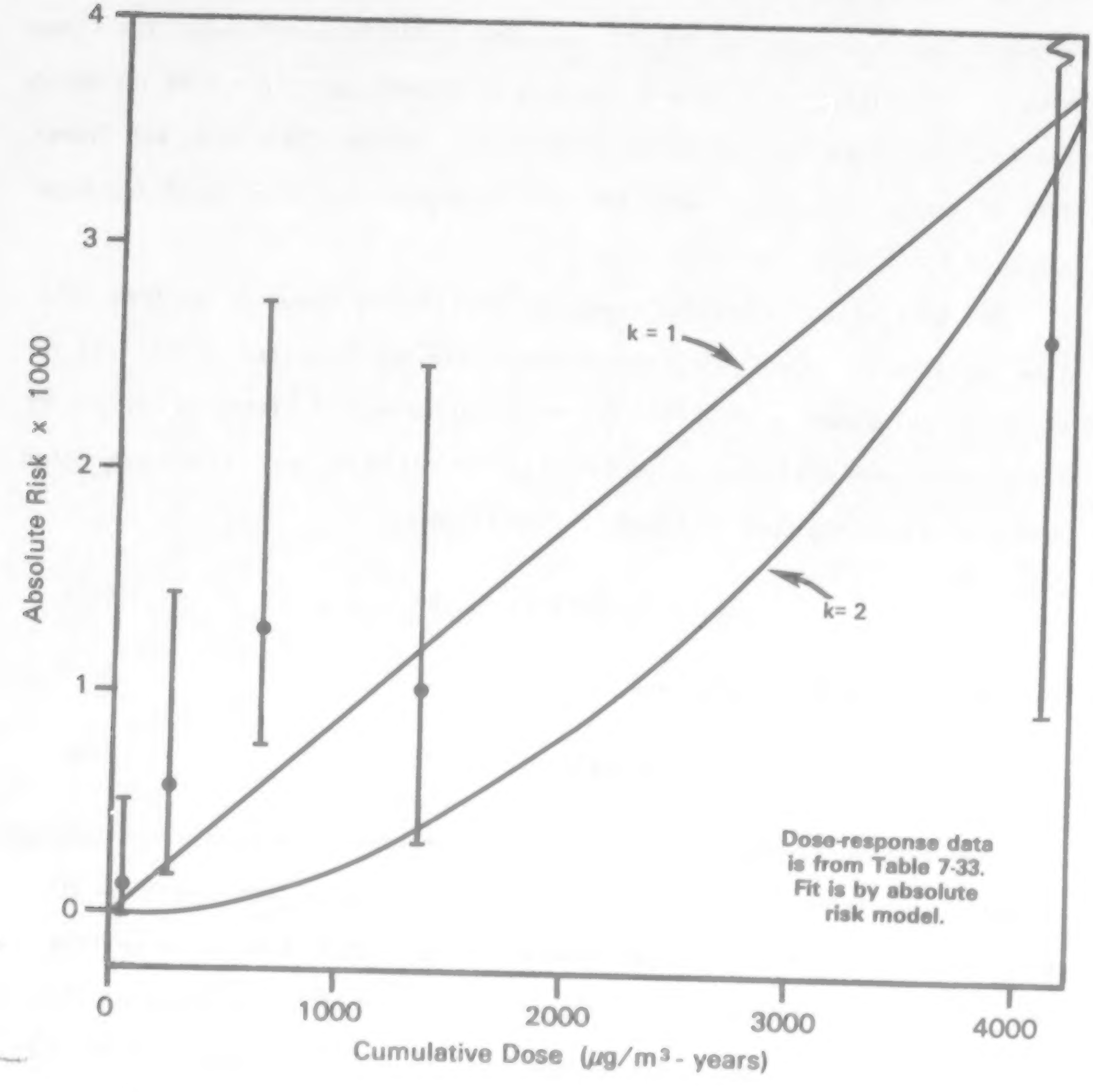


Figure 7-8. Absolute risks and 90% confidence limits for zero-lag data of Enterline and Marsh (1982).



Dose-response data is from Table 7-33. Fit is by absolute risk model.

Figure 7-9. Absolute risks and 90% confidence limits for 10 year lag data of Enterline and Marsh (1982).

potency parameter a' are also listed in Table 7-25. The maximum-likelihood fits of these models are graphed in Figures 7-6 through 7-9. The quadratic fit ($k = 2$) is poor for both the absolute- and relative-risk models with either 0 lag or 10-year lag data (p less than 0.001 in each case). The linear fits to the relative-risk models are also relatively poor ($p = 0.02$ for the 0 lag data and 0.006 for the 10-year lag data). On the other hand, the linear fits of the absolute-risk model are all acceptable ($p = 0.24$ for 0 lag data and 0.14 for 10-year lag data).

The unit risks (additional risks of respiratory cancer death from lifetime exposure to 1 $\mu g/m^3$ airborne arsenic) obtained from each of the fits for which the chi-square p-value is 0.01 or higher, are also listed in Table 7-25. These risks were estimated by applying (1) or (2), (8), and (9) with D_i based upon a constant exposure of 1 $\mu g/m^3$. Specifically,

$$D_i = 4.56[5(i-1) + 2.5] \tag{17}$$

was used for the 0 lag data, and

$$D_i = 4.56[5(i-1) - 7.5] \tag{18}$$

was used for the 10-year lag data. These D_i represent the cumulative exposure in $\mu g/m^3$ resulting from a constant exposure to 1 $\mu g/m^3$ from birth to the midpoint of the ith 5-year age interval. The factor 4.56 converts from occupational to environmental exposures, and is explained at equation (11).

7.3.2.2.5 The Ott et al. (1974) study. Ott et al. (1974) compared the age-specific death patterns of 174 decedents exposed to arsenic in the production of pesticides to those of 1809 decedents who were not exposed to arsenicals. By fitting the death patterns of the unexposed decedents to a mathematical function, an estimate was obtained of the probability that a death at a particular age and during a particular epoch was due to respiratory cancer. This function was used to estimate expected respiratory cancer deaths in various exposure categories for the exposed decedents. Cumulative exposures were estimated for exposed decendents, using work histories and estimates of average exposures in various jobs. The exposure estimates were made by industrial hygienists familiar with the processes. Expected cancer deaths were compared with observed to obtain observed-to-expected ratios. Table 7-34 shows the results of Ott et al.'s dose-response analysis. The data in this table are all reproduced directly from Table 4 of Ott et al. (1974), except for the cumulative exposures. Average total exposures in mg provided by Ott et al. were converted to cumulative exposures in μg/m³ years by multiplying by the factor

$$\frac{1000\ \mu g/mg}{(4\ m^3/day)(21\ days/mo)(12\ mo/year)} \qquad (19)$$

TABLE 7-34. DATA FROM TABLE 4 OF OTT ET AL. (1974)

Cumulative Exposure[a] μg/m³-years	Observed Deaths	Expected Deaths
41.8	1	1.77
125	2	1.01
250	4	1.38
417	3	1.36
790	3	1.70
1544	2	0.97
3505	3	0.77
6451	5	0.79
29497	5	0.72

[a] Exposures are in μg/m³-years estimated by:

$$\frac{d\ mg \times 1000\ \mu g/mg}{(4\ m^3/day)(21\ days/month)(12\ months/year)}$$

where d is average total exposure from Table 4 of Ott et al.

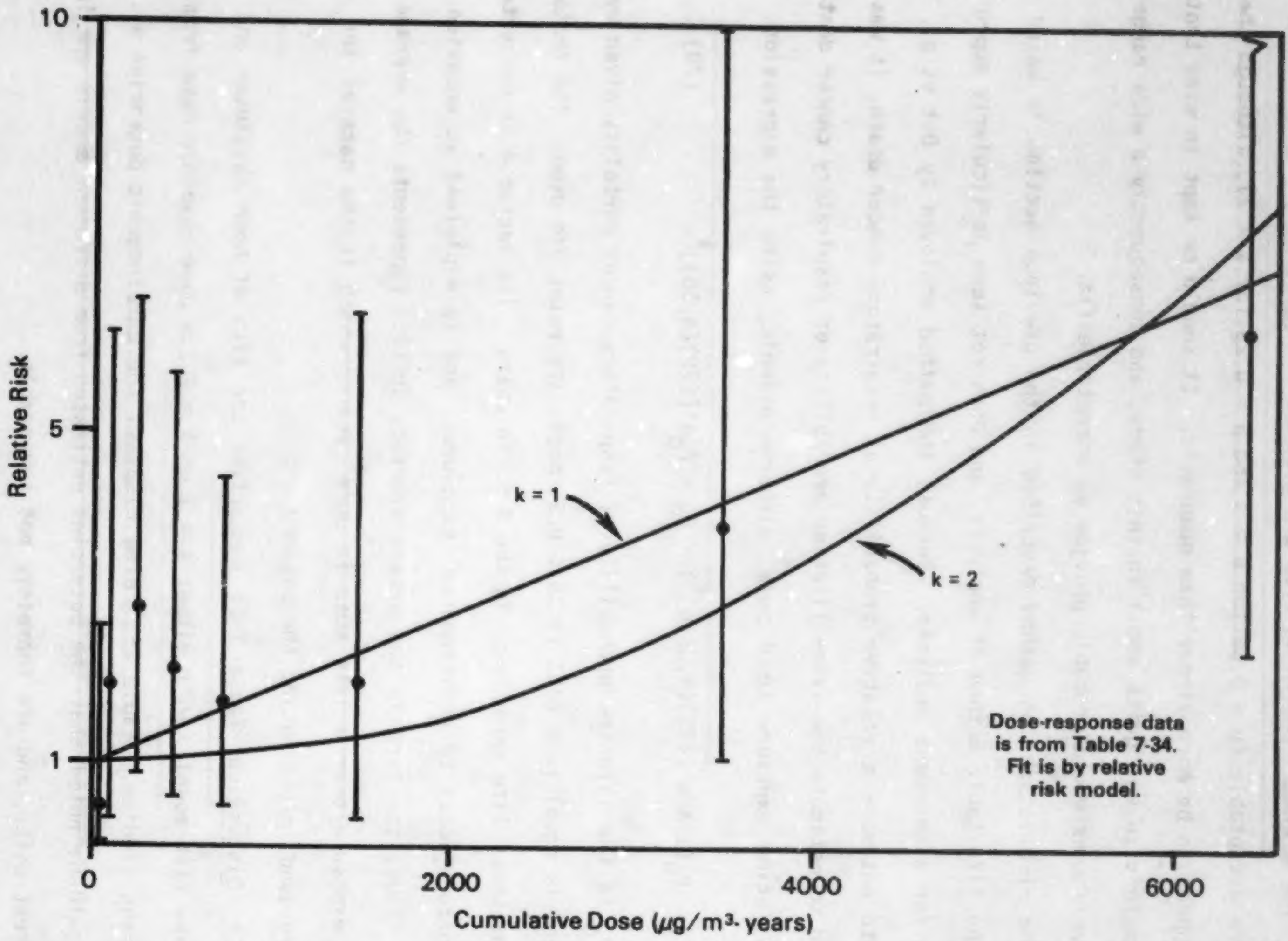
The values included in this factor are not in doubt because use of the factor simply negates the calculation of total exposure made by Ott et al.

Decedent studies such as this are more subject to bias then prospective studies such as those of Lee-Feldstein (1983) and Enterline and Marsh (1982). If, for example, in some age category arsenic exposure increased the mortality from some other disease in addition to respiratory cancer, an analyis of decedents might show an artificially low effect of arsenic upon respiratory cancer for this age group (because there might be an artificially large number of total deaths). It is also of some concern that Ott et al. did not clearly describe how the study cohort was defined.

This study involved primarily short-term exposures, as less than 25 percent of the decedents had worked with arsenicals for more than one year. Thus, this study is less appropriate for estimating risks from lifetime environmental exposure than a comparable study involving longer exposures. The study also was quite small; only 28 respiratory cancer deaths occurred among exposed decedents.

Development of risk estimates: The dose-response data in Table 7-34 were used in an assessment of risk. Because of the nature of the study, only a relative risk model could be applied to these data. The dose-response for relative risk is graphed in Figure 7-10. The response in the most highly exposed group falls far below that predicted by the lower-dose data and is omitted from Figure 7-10. This is possibly due to the fact that some of the more highly exposed workers wore respirators. Because of this shortfall in response, and also because the exposures to this group were the furthest from the low-level environmental exposures of interest, the data from the highest exposure group were omitted from the analysis.

Results of applying chi-square goodness-of-fit tests of the relative-risk model with k = 1 and k = 2 are listed in Table 7-25. The maximum-likelihood



Dose-response data is from Table 7-34. Fit is by relative risk model.

Figure 7-10. Relative risks and 90% confidence limits for data of Ott et al. (1974) with highest exposure group omitted.

estimates of the carcinogenic potency parameter a' are also listed in this table. The maximum-likelihood fits are graphed in Figure 7-10. Both of these fits are acceptable ($p = 0.66$ for $k = 1$ and $p = 0.23$ for $k = 2$), although the data appear to be more linear than quadratic. It should be kept in mind that the sample size was quite small in this study, and consequently a wide range of curve shapes would probably provide an acceptable fit.

The risk estimation method described in the previous section is based upon the life table method of analysis, and does not seem particularly appropriate for a decedent analysis. Because the method employed by Ott et al. seems to estimate a relative probability of respiratory cancer death, it was decided to estimate the extra lifetime probability of respiratory cancer death from lifetime exposure to d $\mu g/m^3$ airborne arsenic, using the expression

$$P_0(1 + a'[(72)(4.56)d]^k) - P_0 = P_0 a'[(72)(4.56)]^k. \qquad (20)$$

Here P_0 is the lifetime probability of respiratory cancer mortality given by (8), and is equal to 0.0451 if 1976 U.S. mortality rates are used. The factor 72 represents life expectancy in the U.S. in years. The factor 4.56 converts from occupational to environmental exposures, and is explained at equation (11). Thus, the term in the square brackets in (20) represents the average total exposure over a life span in $\mu g/m^3$ years, which is the same as the measure used in estimating the potency a'.

7.3.2.3 Discussion--Table 7-25 summarizes the fits of both absolute- and relative-risk models, with either $k = 1$ or $k = 2$, to dose-response data from 4 different studies. Table 7-25 also displays the carcinogenic potencies a'. It should be noted that the potencies estimated from different models are in different units, and are therefore not comparable.

In every case, a linear model ($k = 1$) fitted the data better than the corresponding quadratic model ($k = 2$). In every case but two, the fits of the quadratic model could be rejected at the 0.01 level. The two exceptions involved the two smallest data sets (Higgins et al. absolute risk, and Ott et al.) and in the former case the fit was very marginal ($p = 0.017$). On the other hand, for each data set a linear model provided an adequate fit. Also, in every case, an absolute-risk linear model fit the data better than the corresponding relative-risk linear model. The p-values for the fits of the absolute-risk linear model ranged from 0.025 to 0.75.

The estimated unit risk is presented for each fit for which the chi-square goodness-of-fit p-value is greater than 0.01. The unit risks derived from linear models--8 in all--range from 0.0013 to 0.0136. The largest of these is from the Ott et al. study, which probably is the least reliable for developing quantitative estimates, and which also involved exposures to pentavalent arsenic, whereas the other studies involved trivalent arsenic. The unit risks derived from the linear ($k = 1$) absolute-risk models are considered to be the most reliable; although derived from 5 sets of data involving 4 sets of investigators and 2 distinct exposed populations, these estimates are quite consistent, ranging from 0.0013 to 0.0076.

To establish a single point estimate, the geometric mean for data sets is obtained within distinct exposed populations, and the final estimate is taken to be the geometric mean of those values. This process is illustrated in Table 7-35.

The final estimate is 4.29×10^{-3}, where exposure is in $\mu g/m^3$ of continuous exposure. Based upon an assumed 20 m^3 tidal volume of air and a 30 percent absorption rate, this amounts to a unit risk of $4.29 \times 10^{-3} \div (.3 \times 20 \times .001/70) = 50.1$ in units of mg/kg absorbed dose per day.

Although the estimates derived from the various studies are quite consistent, there are a number of uncertainties associated with them. The estimates

TABLE 7-35. COMBINED UNIT RISK ESTIMATES FOR ABSOLUTE-RISK LINEAR MODELS

Exposure Source	Study	Unit Risk	Geometric Mean Unit Risk	Final Estimated Unit Risk
Anaconda smelter	Brown & Chu	1.25×10^{-3}	2.56×10^{-3}	4.29×10^{-3}
	Lee-Feldstein	2.80×10^{-3}		
	Higgins	4.90×10^{-3}		
ASARCO smelter	Enterline & Marsh	6.81×10^{-3}	7.19×10^{-3}	
		7.60×10^{-3}		

were made from occupational studies that involved exposures only after employment age was reached. In estimating risks from environmental exposures throughout life, it was assumed, through either the relative-risk model (1) or the absolute-risk model (2), that the increase in the age-specific mortality rates of lung cancer was a function only of cumulative exposures, irrespective of how the exposure was accumulated. Although this assumption provides an adequate description of all of the data, it may be in error when applied to exposures that begin very early in life. Similarly, the linear models possibly are inaccurate at low exposures, even though they provide excellent descriptions of the experimental data.

The risk assessment methods employed were severely constrained by the fact that they were based only upon the analyses performed and reported by the original authors--analyses that had been performed for purposes other than quantitative risk assessment. For example, although other measures of exposure might be more appropriate, the analyses were necessarily based upon cumulative dose, since that was the only usable measure reported. Given greater access to the data from these studies, other dose measures, as well as models other than the simple relative-risk and absolute-risk models, could be studied. It is possible that such wide analyses would indicate that other approaches are more appropriate than the ones applied here.

7.3.3 Unit Risk for Water

The best data available for making quantitative cancer risk estimates for ingestion of arsenic in water are the data collected by Tseng et al. (1968). They surveyed a stable population of 40,421 individuals who lived in a rural area along the southwest coast of Taiwan and who were known to have consumed drinking water containing arsenic. The occurrences of skin cancer among this population, and the arsenic concentrations in their drinking water, were measured. Since the population was stable, the study can be viewed as a lifetime feeding study, and the data may be used to predict the lifetime probability of skin cancer caused by the ingestion of arsenic.

A model estimating the cancer rate as a function of drinking water arsenic concentration was generated using information from the above study in its published form, which is a summary of data collected by the investigators. If the original data had been available, a more exact mathematical analysis would have been possible.

Doll (1971) has suggested that the relationship between the incidence of some site-specific cancers, age, and exposure level of a population may be expressed as:

$$I(x,t) = kBx^{m}t^{k-1} \qquad (21)$$

where x is the exposure level (which can be measured by the water concentration in ppm), t is the age of the population, and B, m, k are unknown parameters.

However, the data collected by Tseng et al. (1968) was obtained at one point in time, and since skin cancer has only a marginal effect on the death rate, the obtained rates may be viewed more accurately as the probability of having contracted skin cancer by time t. The relationship between this probability, often referred to as the cumulative probability density or prevalence F(x,t), and the incidence or age-specific or hazard rate, may be expressed as:

$$F(x,t) = 1 - \exp\left[-\int_o^t I(x,s)\,ds\right]. \qquad (22)$$

Utilizing equation (21) as the form of the incidence rate, the prevalence may be expressed as

$$F(x,t) = 1 - \exp\left(-Bx^m t^k\right), \qquad (23)$$

which is a Weibull distribution.

In Table 7-36, adapted from information in Tseng, et al. (1968), estimates are given of F(x,t) for different age and water concentration groupings for males. The prevalence for females is less than for males, and therefore is not used to estimate risk.

To use this data, specific values for x and t had to be obtained for the intervals. Where the intervals were closed, the midpoint was utilized. For the greater than 0.6 ppm group, the midpoint between 0.6 and the greatest recorded value, 1.8, was taken, resulting in 1.2 ppm. For age 60 or greater, a value of 70 was utilized somewhat arbitrarily, being the same increase over the lower level as that in the other two age intervals. The values for (x,t) to relate to the prevalence estimates are shown in parentheses in Table 7-36.

From equation (23) it follows that

$$\ln(-\ln[1-F(x,t)]) = \ln(B) + m\ln(x) + k\ln(t), \qquad (24)$$

which is multiple-linear in form. Estimating the parameters by the usual least-square techniques, the following relationship is obtained:

TABLE 7-36. AGE-EXPOSURE-SPECIFIC PREVALENCE RATES FOR SKIN CANCER

	AGE		
Exposure in ppm[a]	20-39 (30)	40-59 (50)	≥60 (70)
0 - 0.29 (0.15)	0.0013	0.0065	0.0481
0.30 - 0.59 (0.450)	0.0043	0.0477	0.1634
≥0.6 (1.2)	0.0224	0.0983	0.2553

*Source: Tseng et al. (1968).

[a] Range given by authors. Midpoint is in parentheses.

$$\ln(-\ln[1 - F(x,t)]) = 17.548 + 1.192 \ln(x) + 3.881 \ln(t), \quad (25)$$

which is an excellent fit, having a multiple correlation coefficient of 0.986 and a standard error on the exposure regression m of .138.

Equation (25) may be expressed as

$$\begin{aligned} F(x,t) &= 1\text{-}\exp\text{-}[2.429 \times 10^{-8}(X^{1.192})(t^{3.881})] \\ &= 1\text{-}\exp\text{-}[X^{1.192} H(t)] \qquad (26) \end{aligned}$$

If the parameter m = 1.192 were in fact equal to 1, then for a given value of t, equation (26) would be "one-hit" in form.

To test this hypothesis (i.e., Ho: m = 1) the student "t" test is used, giving the result:

$$t_6 = \frac{1.192 - 1}{0.138} = 1.391,$$

which is not significant at the 0.1 level. Thus, there is insufficient evidence to reject the hypothesis that the dose-response relationship is "one-hit" even at the 0.1 level. However, a quadratic model would be rejected at the $p<.001$ level.

Fixing $m = 1$, the following relationship is obtained:

$$F(x,t) = 1 - \exp[-g(t)x] \qquad (27)$$

Transforming this equation to its linear form (as in equation 23) and obtaining the least-square linear estimates of B and v, it is found that:

$$g(t) = \exp(-17.5393)\, t^{3.853}, \text{ where } B = 2.41423 \times 10^{-8},\ k = 3.853.$$

The data used to obtain these estimates are shown in Table 7-37, and the goodness-of-fit is illustrated in Figure 7-11.

TABLE 7-37. DATA UTILIZED TO OBTAIN PREDICTOR EQUATION AND FIGURE 7-11

ppm Arsenic	Age at Medical Examination	Skin Cancer Prevalence Rate		Transformed Skin Cancer Prevalence Rate	
		$F(x,t)$		$-\ln(-\ln[1-F(x,t)]) = -17.5393 + 3.853\ln t + \ln x$	
x	t	Observed Rate	Expected Rate	Observed	Expected
0.15	30	0.0013	0.0031	6.64474	6.33160
	50	0.0063	0.0127	5.03269	4.36341
	70	0.0481	0.0455	3.00993	3.06695
0.45	30	0.0043	0.0053	5.44699	5.23299
	50	0.0477	0.0375	3.01849	3.26480
	70	0.1634	0.1304	1.72368	1.96834
1.20	30	0.0224	0.0141	3.78739	4.25216
	50	0.0983	0.0969	2.26844	2.28397
	70	0.2553	0.3110	1.22155	0.98751

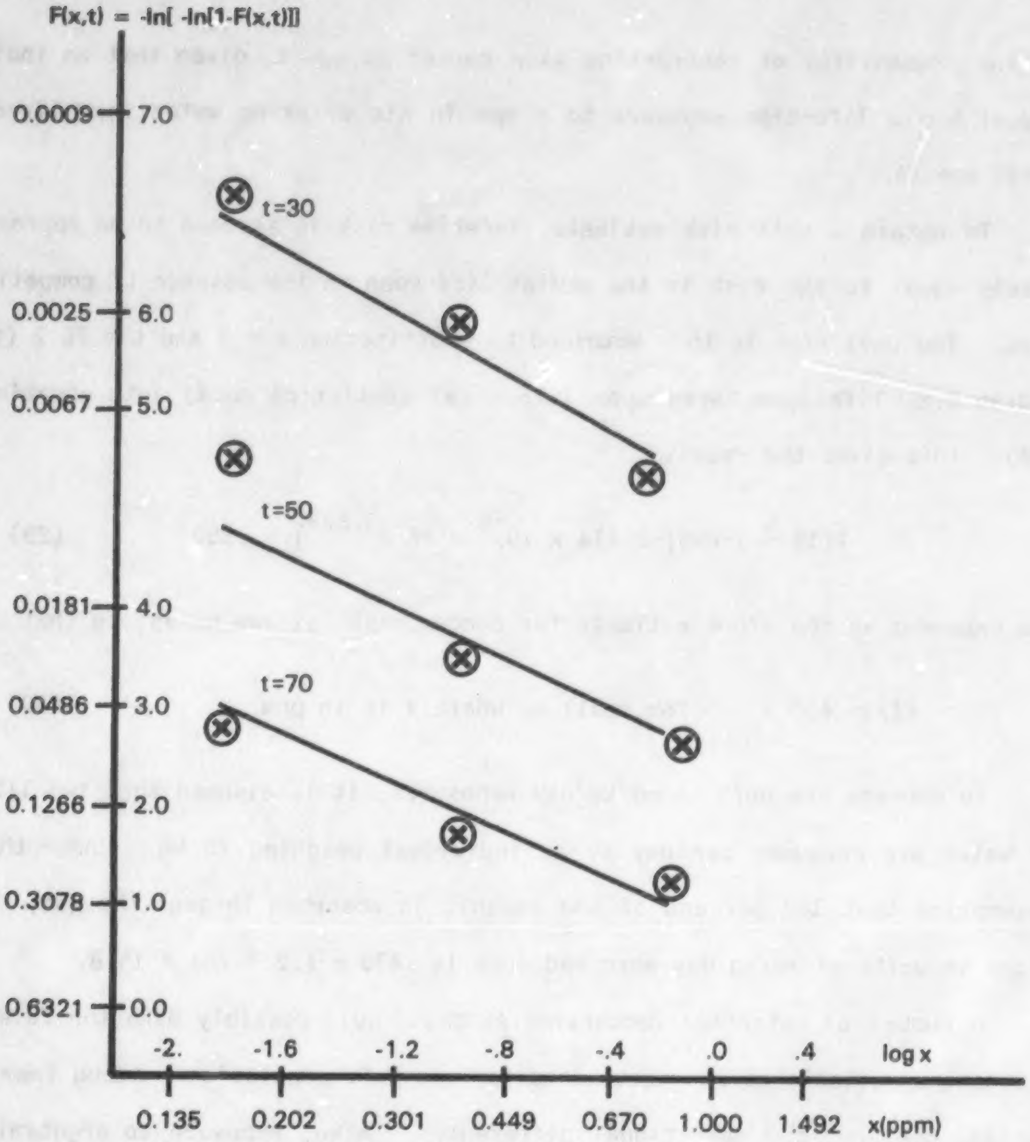


Figure 7-11. Relationship between transformed prevalence and log ppm arsenic in water, log age.

The function

$$F(x,t) = 1\text{-}\exp[-2.41423 \times 10^{-8} \times t^{3.853}], \qquad (28)$$

is the probability of contracting skin cancer by age t, given that an individual had a life-time exposure to x ppm in his drinking water (and lived until age t).

To obtain a unit risk estimate, lifetime risk is assumed to be approximately equal to the risk to the median life span in the absence of competing risk. The unit risk is thus obtained by substituting $x = 1$ and $t = 76.2$ (the median U.S. life span based upon 1976 vital statistics data) into equation (28). This gives the result

$$P(1) \simeq 1\text{-}\exp[-2.414 \times 10^{-8} \times 76.2^{3.853}] = .350 \qquad (29)$$

The exponent is the slope estimate for cancer risks at low doses, so that:

$$P(x) \simeq .430\,x \quad \text{for small } x \text{, where } x \text{ is in ppm.} \qquad (30)$$

To express the unit in mg/kg/day exposures, it is assumed that two liters of water are consumed per day by an individual weighing 70 kg. Under the assumption that 100 percent of the arsenic is absorbed through the gut, the slope in units of mg/kg/day absorbed dose is $.430 \div (.2 \div 70) = 15.8$.

A number of potential factors exist that could possibly make the Taiwanese data unsuitable as surrogate data for the U.S. population. Among them are racial, dietary, and nutritional differences. Also, exposure to ergotamine was confounded with arsenic exposure in the well water--a fact which also could have modified the results. However, there is no direct evidence demonstrating the role of these agents in the carcinogenic response to ingested arsenic. Furthermore, a recent extensive review by Andelman and Barnett (1983) of the arsenic dose-response model developed here, demonstrates that presently there is no quantitative evidence that is inconsistent with the model. The Andelman and Barnett study also showed that there does not appear to be any population in the U.S. that could be studied that would have a reasonable power to contradict the hypothesis that the Taiwanese dose-response model is consistent with the U.S. dose response.

7.3.4 Relative Potency

One of the uses of the concept of unit risk is to compare the relative potencies of carcinogens. To estimate relative potency on a per-mole basis, the unit risk slope factor is multiplied by the molecular weight, and the resulting number is expressed in terms of $(\text{mMol/kg/day})^{-1}$. This is called the relative potency index.

Figure 7-12 is a histogram representing the frequency distribution of potency indices of 52 chemicals evaluated by the CAG as suspect carcinogens. The actual data summarized by the histogram are presented in Table 7-38. Where human data were available for a compound, they were used to calculate the index. Where no human data were available, animal oral studies and animal inhalation studies were used, in that order. Animal oral studies were selected over animal inhalation studies because they have been made on most of the chemicals, thus allowing potency comparisons by route.

The potency index for arsenic, based on the Tseng et al. study, is 2.25×10^3 $(\text{mMol/kg/day})^{-1}$. This is derived by means of the slope estimate from the Tseng et al. study, which is $15(\text{mg/kg/day})^{-1}$.

Multiplication by the molecular weight of 149.8 gives a potency index of $2.25 \times 10^{+3}$. Rounding off to the nearest order of magnitude gives a value of 10^{+3}, which is the scale presented on the horizontal axis of Figure 7-12. The index of $2.25 \times 10^{+3}$ lies at the bottom of the first quartile of the 52 suspect carcinogens.

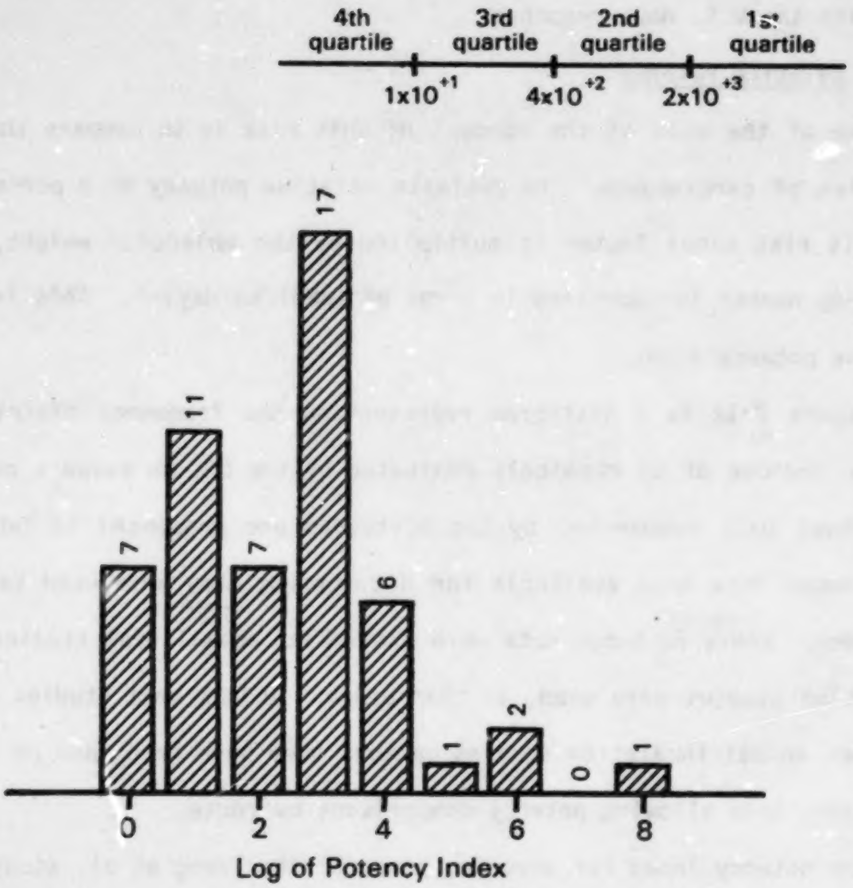


Figure 7-12. Histogram representing the frequency distribution of the potency indices of 52 suspect carcinogens evaluated by the Carcinogen Assessment Group.

TABLE 7-38. RELATIVE CARCINOGENIC POTENCIES AMONG 52 CHEMICALS EVALUATED BY THE CARCINOGEN ASSESSMENT GROUP AS SUSPECT HUMAN CARCINOGENS

Compounds	Slope (mg/kg/day)-1	Molecular Weight	Potency Index	Order of Magnitude (log$_{10}$) Index
Acrylonitrile	0.24(W)	53.1	$1 \times 10^{+1}$	+1
Aflatoxin B_1	2924	312.3	$9 \times 10^{+5}$	+6
Aldrin	11.4	369.4	$4 \times 10^{+3}$	+4
Allyl Chloride	1.19×10^{-2}	76.5	9×10^{-1}	0
Arsenic	15(H)	149.8	$2 \times 10^{+3}$	+3
B[a]P	11.5	252.3	$3 \times 10^{+3}$	+3
Benzene	5.2×10^{-2}(W)	78	4×10^{0}	+1
Benzidine	234(W)	184.2	$4 \times 10^{+4}$	+5
Beryllium	4.86	9	$4 \times 10^{+1}$	+2
Cadmium	6.65(W)	112.4	$7 \times 10^{+2}$	+3
Carbon Tetrachloride	1.30×10^{-1}	153.8	$2 \times 10^{+1}$	+1
Chlordane	1.61	409.8	$7 \times 10^{+2}$	+3
Chlorinated Ethanes				
1,2-dichloroethane	6.90×10^{-2}	98.9	7×10^{0}	+1
1,1,2-trichloroethane	5.73×10^{-2}	133.4	8×10^{0}	+1
1,1,2,2-tetrachloroethane	0.20	167.9	$3 \times 10^{+1}$	+1
Hexachloroethane	1.42×10^{-2}	236.7	3×10^{0}	0
Chloroform	7×10^{-2}	119.4	8×10^{0}	+1
Chromium	41	104	$4 \times 10^{+3}$	+4
DDT	8.42	354.5	$3 \times 10^{+3}$	+3
Dichlorobenzidine	1.69	253.1	$4 \times 10^{+2}$	+3
1,1-dichloroethylene	1.04(I)	97	$1 \times 10^{+2}$	+2
Dieldrin	30.4	380.9	$1 \times 10^{+4}$	+4

TABLE 7-38. (continued)

Compounds	Slope (mg/kg/day)-1	Molecular Weight	Potency Index	Order of Magnitude ($\log_{10}$) Index
Dinitrotoluene	0.31	182	$6x10^{+1}$	+2
Diphenylhydrazine	0.77	180	$1x10^{+2}$	+2
Epichlorohydrin	$9.9x10^{-3}$	92.5	$9x10^{-1}$	0
Bis(2-chloroethyl)ether	1.14	143	$2x10^{+2}$	+2
Bis(chloromethyl)ether	9300(I)	115	$1x10^{+6}$	+6
Ethylene Dibromide (EDB)	8.51	187.9	$2x10^{+3}$	+3
Ethylene Oxide	0.63(I)	44.0	$3x10^{+1}$	+1
Formaldehyde	$2.14x10^{-2}$(I)	30	$6x10^{-1}$	0
Heptachlor	3.37	373.3	$1x10^{+3}$	+3
Hexachlorobenzene	1.67	284.4	$5x10^{+2}$	+3
Hexachlorobutadiene	$7.75x10^{-2}$	261	$2x10^{+1}$	+1
Hexachlorocyclohexane				
technical grade	4.75	290.9	$1x10^{+3}$	+3
alpha isomer	11.12	290.9	$3x10^{+3}$	+3
beta isomer	1.84	290.9	$5x10^{+2}$	+3
gamma isomer	1.33	290.9	$4x10^{+2}$	+3
Nickel	1.15(W)	58.7	$7x10^{+1}$	+2
Nitrosamines				
Dimethylnitrosamine	25.9(not by q_1^*)	74.1	$2x10^{+3}$	+3
Diethylnitrosamine	43.5(not by q_1^*)	102.1	$4x10^{+3}$	+4
Dibutylnitrosamine	5.43	158.2	$9x10^{+2}$	+3
N-nitrosopyrrolidine	2.13	100.2	$2x10^{+2}$	+2
N-nitroso-N-ethylurea	32.9	117.1	$4x10^{+3}$	+4
N-nitroso-N-methylurea	302.6	103.1	$3x10^{+4}$	+4
N-nitroso-diphenylamine	$4.92x10^{-3}$	198	$1x10^{0}$	0
PCBs	4.34	324	$1x10^{+3}$	+3
Phenols				
2,4,6-trichlorophenol	$1.99x10^{-2}$	197.4	4x10	+1

TABLE 7-38. (continued)

Compounds	Slope (mg/kg/day)-1	Molecular Weight	Potency Index	Order of Magnitude ($\log_{10}$) Index
Tetrachlorodioxin	4.25×10^{5}	322	$1\times10^{+8}$	+8
Tetrachloroethylene	5.31×10^{-2}	165.8	9×10^{0}	+1
Toxaphene	1.13	414	$5\times10^{+2}$	+3
Trichloroethylene	1.26×10^{-2}	131.4	2×10^{0}	0
Vinyl Chloride	1.75×10^{-2}(I)	62.5	1×10^{0}	0

Remarks:

1. Animal slopes are 95% upperlimit slopes based on the linear multistage model. They are calculated based on animal oral studies, except for those indicated by I (animal inhalation), W (human occupational exposure), and H (human drinking water exposure). Human slopes are point estimate, based on the linear non-threshold model.
2. The potency index is a rounded-off slope in (mMol/kg/day)-1 and is calculated by multiplying the slopes in (mg/kg/day)-1 by the molecular weight of the compound.
3. Not all the carcinogenic potencies presented in this table represent the same degree of certainty. All are subject to change as new evidence becomes available.

Ranking of the relative potency indices is subject to the uncertainty involved in comparing estimates of potency for different chemicals based on different routes of exposure to different species, and using studies of different quality. Furthermore, all the indices are based on estimates of low-dose risk using linear extrapolation from the observational range. Thus, these indices are not valid for the purpose of comparing potencies in the experimental or observational range if linearity does not exist there.

7.3.5 Summary and Conclusions of the Carcinogenicity of Arsenic

7.3.5.1 Qualitative Summary--Human studies of the effects of arsenic from smelters, drinking water, pesticide manufacturing plants, and medicinals have been conducted. These are summarized in Table 7-1. Studies of five independent smelter worker populations have all found an association between occupational arsenic exposure and lung cancer mortality. Several of the smelter studies have found a dose response both by intensity and by duration of exposure. The risk of lung cancer mortality in the high dose group of one study of smelter workers in Japan was found to be 10 times that expected. Some of the recent data from these smelter studies suggest that, if the multistage theory of carcinogenesis is correct, arsenic may act as a late-stage carcinogen. It is also possible to hypothesize from some of the data that arsenic may be a promoter. Either of these hypotheses, however, requires further study. In addition, some studies of communities surrounding smelters have found an association between geographic proximity to the smelter and lung cancer mortality.

Both proportionate mortality and cohort studies of pesticide manufacturing workers have demonstrated an excess of lung cancer deaths among workers in that occupation. One study of the population around a pesticide manufacturing plant found that residents of the area surrounding the plant were also at an excess risk of lung cancer. Several case reports of arsenical pesticide applicators have also shown an association between arsenical exposure and lung cancer.

A study of 40,000 persons in Taiwan exposed to arsenic in the drinking water found a significant excess prevalence of skin cancer over that of 7,500 other Taiwanese and residents of Matsu Island who drank water relatively free of arsenic. Water supplies in Chile and Argentina were also reported to be the cause of arsenic-induced skin cancers. In contrast, studies of populations in the United States exposed to relatively high levels of arsenic in the drinking water by U.S. standards did not find any excess of skin cancer.

The difference in response between the Taiwanese and the United States studies may reflect the fact that the drinking water in the study area in Taiwan contained much higher levels of arsenic than did the drinking water in the United States. Furthermore, the Taiwanese waters also contained ergotamine-like compounds. Other differences between the two study areas include differences in socioeconomic status, with the Taiwanese who developed skin cancer belonging to a rather low socioeconomic strata which could have had ramifications in terms of diet, personal hygiene and medical care. Racial differences may have also contributed to the differences between the study results. In addition to these physical and cultural differences, the U.S. studies were limited by small sample sizes and would not have detected the risk from skin cancer that would have been predicted, on the basis of arsenic ingestion, from a linear model of the Taiwanese data.

Persons exposed to arsenical medicinals have also been shown to be at a risk of skin cancer.

Using the International Agency for Research on Cancer (IARC) classification scheme for evaluating carcinogens, the evidence for arsenic as a human carcinogen is considered sufficient. This is evidenced by the high relative risks, the consistency in findings in different studies, and the specificity of tumor sites (i.e., skin and lungs).

Consistent demonstration of arsenic carcinogenicity in test animals, using different chemical forms, routes of exposure, and various experimental species, has not been observed. Nevertheless, recent data indicate that tumorigenicity and possibly carcinogenicity can be demonstrated in animals if the retention of arsenic in the lung is increased. The additional observation that calcium arsenate is only slowly cleared from the lung, strongly suggests that this agent may be carcinogenic. In support of the recent animal studies, is the observation that cultured Syrian hamster embryo cells can be transformed by direct exposure to sodium arsenate.

7.3.5.2 Quantitative summary--Unit risks are estimated for both air and water exposures to arsenic. The air estimates were based on data obtained in five separate studies involving three independently exposed worker populations. Linear and quadratic dose-response models in both the absolute and relative form are fitted to the worker data. It was found that for the models that fit the data at the P = .01 or better level that the corresponding unit risk estimates ranged from 1.05×10^{-4} to 1.36×10^{-2}. However, linear models fit better than quadratic models and absolute better than relative models. Also it was felt that exposure to trivalent arsenic was more representive of low environmental exposure than pentavalent arsenic. Restricting unit risk estimates to those obtained from linear absolute models where exposure was to trivalent arsenic gives a range of 1.25×10^{-3} to 7.6×10^{-3}. A weighted average of the five estimates in this range gave a composite estimate of 4.29×10^{-3}.

An extensive drinking water study of the association between arsenic in well water and an examination for skin cancer of a population who lived in a rural area of Taiwan was used to estimate the unit risk for ingestion. Using the male population who appeared to be more susceptible, it was estimated that the unit risk associated with drinking water contaminated with 1 μg/ℓ of arsenic was 4.3×10^{-4}. To compare the air and water unit risks, the exposure units in both cases were converted to mg/kg/day absorbed doses, resulting in unit risk estimates of 50.1 and 15.0, respectively.

The potency of arsenic compared to other carcinogens is evaluated by noting that an arsenic potency of $2.25 \times 10^{+3}$ $(\text{mMol/kg/day})^{-1}$ lies in the first quartile of the 52 suspect carcinogens that have been evaluated by CAG.

7.3.5.3 Conclusions--Skin cancer and lung cancer have been shown by numerous epidemiologic studies to have an association with arsenic exposure. Arsenic has not definitively been found to be a carcinogen in animal studies, however. In applying the IARC criteria for evaluating a substance as to the weight of evidence for human carcinogenicity, arsenic would be placed in group 1, which IARC characterizes as "carcinogenic to humans."

Using the linear absolute risk model, the composite estimate for cancer risk due to a lifetime exposure to 1 $\mu g/m^3$ trivalent arsenic in the air is estimated to be 4.29×10^{-3}. The unit risk due to lifetime exposure to 1 $\mu g/\ell$ of arsenic in drinking water is estimated to be 4.3×10^{-4}. On the basis of mg/kg/day absorbed dose, the unit-risk slope estimates for air and water are 50.1 and 15, respectively. While it is unlikely that the true risks would be higher than these estimates, they could be substantially lower. Compared to other compounds on a mole unit basis, the carcinogenic potency for arsenic falls towards the lower end of the first quartile.

8. ARSENIC AS AN ESSENTIAL ELEMENT

Mertz (1970) has set forth a set of logical criteria which trace elements should obey in order to be considered physiologically essential for man and/or animals. One of the most obvious of these, and one which should be readily demonstrable, is that the element meet a unique requirement physiologically, and, consequently, that a deficiency in that element be associated with deleterious effects.

In the case of arsenic, early reports attempting to show a nutritional requirement for the element in animals were inconclusive (Arsenic. NAS, 1977; Underwood, 1977). Part of the problem was undoubtedly technical in nature, i.e., the difficulty of carrying out such studies in an experimental environment where rigorous exclusion of a ubiquitous element from the diet is necessary. More recently, however, several carefully controlled studies have been reported to have demonstrated nutritional essentiality for arsenic in at least some mammalian species.

Nielsen et al. (1978) have noted that deprivation of pregnant rats of arsenic-supplemented diets resulted in offspring showing such post-weaning effects as slow growth, enlarged spleens, and increased red cell osmotic fragility. Greater perinatal mortality among pups from arsenic-deprived dams was also noted in a second experimental group.

In a recent review by Uthus et al. (1983), the authors reported on studies with chicks that suggest that arsenic influences arginine metabolism. It was reported that arsenic deprivation influenced the effects of dietary arginine, manganese and zinc, the fluctuations of which variously affected growth, kidney arginase, plasma alkaline phosphatase, plasma urea, plasma uric acid and hematocrit. The authors suggested that the four components interacted to affect the conversion of arginine into urea and ornithine.

Anke et al. (1978) studied the nutritional requirements for arsenic using goats and mini-pigs and a semi-synthetic diet containing less that 50 ppb arsenic. Effects attributed to arsenic deficiency in both species were seen not only in the adult animals but in their offspring. Arsenic deficiency increased the mortality of adult goats as well as altered the mineral profiles for copper and manganese in the carcass. Significant reproductive effects for both arsenic-deficient goats and mini-pigs included reduction of the normal litter size. Furthermore, the mortality of kids and piglets from the arsenic-deficient groups was significantly higher than controls. Manganese levels were elevated in arsenic-deficient kids and piglets, but no perturbation of hematological indices (hemoglobin, hematocrit or mean corpuscular concentration) was noted. This is in contrast to the experimental observations with rats (Nielsen et al., 1974), where decreased hematocrits, elevated iron content in spleen and increased osmotic fragility of cells are seen. Given the fact that the rat is known to be an anomalous animal model for arsenic metabolism (see Chapter 4), this difference is probably peculiar to this species.

Schwartz (1977) has noted growth effects of arsenite on rats fed an arsenic-supplemented diet, with an optimal effect seen at 0.25 to 0.5 ppm. Interestingly, this worker noted that pentavalent arsenic as sodium arsenate is less effective.

Remaining to be independently demonstrated is a physiological role for arsenic, the existence of any specific carrier agent in the body, or arsenic essentiality in man.

A feature of essential element metabolism is homeostatic control of levels and movement of a particular element in vivo. From the information considered earlier, there is no effective absorption barrier for most soluble inorganic arsenicals, but efficient excretory mechanisms (kidney, hair) and biotransformation appear to provide some control over any absorption-excretion balance.

9. HUMAN HEALTH RISK ASSESSMENT FOR ARSENIC

This portion of the report places the information in the earlier Chapters into a quantitative perspective with regard to non-occupational population exposure and health effects of arsenic germane to such a population.

Data for levels of arsenic encountered by humans in air, water, food and other sources, such as cigarette smoking, were set forth in Chapter 3 and are combined with data on rates of intake and rates of absorption to provide information on the total assimilation of arsenic on a daily basis. Health effects of arsenic most germane to non-occupational population exposures are then summarized. Generally, these are chronic effects associated with long-term intake of relatively low levels of arsenic. In the case of hazardous wastes, however, some health effects of concern may be associated with acute exposures; therefore, acute and sub-acute effects must also be considered.

The section dealing with dose-effect/dose-response data includes consideration of various indices of internal exposure followed by quantitative data for intake and population response.

Populations at risk, identified at least along qualitative lines, are included for discussion.

9.1 AGGREGATE EXPOSURE LEVELS TO ARSENIC IN THE U.S. POPULATION

Among individuals of the general population (not occupationally exposed to arsenic), the main routes of exposure to arsenic are typically via ingestion of food and water, with lesser exposures occurring via inhalation. Representative intake figures are presented in Table 9-1. Intake by inhalation is augmented among smokers in proportion to the level of smoking.

TABLE 9-1. ROUTES OF DAILY HUMAN ARSENIC INTAKE

Route/level	Rate	Total intake	Absorbed amount
Ambient air/0.006 μg/m^3[a]	20 m^3	0.12 μg	0.036 μg[b]
Drinking water/≤ 10 μg/liter	2 liters	≤ 20 μg	≤ 20 μg[c]
Food/50 μg daily (elemental As)	--	50 μg	40 μg[d]
Cigarettes/6 μg in main-stream smoke/pack[e]	1/2 pack	3 μg	0.9 μg[f]
	1 pack	6 μg	1.8 μg[f]
	2 pack	12 μg	2.7 μg[f]
Total: ≤ 60 μg nonsmokers			

[a] National average for 1981 (see Section 3.3.1)

[b] Assumes 30 percent respiratory absorption (see Chapter 4).

[c] Assumes total absorption (see Chapter 4).

[d] Assumes 80 percent absorption (see Chapter 4).

[e] Assumes 20 percent of cigarette content in inhaled smoke (see Chapter 4).

[f] Assumes 30 percent absorption of inhaled amount (see Chapter 4).

Assuming a daily ventilation rate of 20 m^3, and a national population inhalation average of 0.006 μg/m^3/As, the total daily inhalation exposure for arsenic can be projected to be approximately 0.12 μg. Assuming 30 percent absorption, approximately 0.03 μg of arsenic would be absorbed on a daily average.

Contribution of tobacco-borne arsenic to the respiratory burden would depend upon the rate of cigarette smoking. If one assumes a mass of 1 gram/cigarette and an average tobacco value of 1.5 ppm, this yields 1.5 μg arsenic/cigarette. With 20 percent of this amount in mainstream smoke, the inhaled amount for each pack of cigarettes would be approximately 6 μg arsenic, and of this amount, 40 percent would be deposited in the respiratory tract (see Chapter 4). Assuming an absorption of 75 percent of the deposited fraction, one arrives at an absorption of approximately 2 μg/pack of cigarettes or a factor of 10 to 100 times greater than intake for nonsmokers in given ambient air settings. One may assume that the rates of absorption for trivalent and pentavalent arsenic in the respiratory tract are equivalent.

Since drinking water arsenic is mainly in a soluble form (arsenate or arsenite), virtually all of it is absorbed in the GI tract (see Chapter 4). Thus, assuming an average daily consumption of two liters of water containing at most 10 µg As/liter as an outside high figure, one can estimate that the total arsenic absorbed from drinking water would be approximately 20 µg/day. Most individuals would, in reality, take in much less than this amount, while those in the Western U.S. with well water supplies much higher in arsenic content would assimilate proportionately more.

Food arsenic values taken from the 1974 FDA survey indicate a daily total dietary intake of approximately 50 µg elemental arsenic. Based on information presented in Chapter 4, the major portion (80 percent) of food arsenic would be absorbed resulting in a net daily food arsenic absorption of 40 µg total.

Thus, a non-smoker would have a total daily absorption from all exposure media of approximately 60 µg arsenic/day or less. Of this, the diet would be the major contributor, assuming levels in water much below 10 µg/liter. For cigarette smokers, one would add 2 µg arsenic/pack of cigarettes smoked daily.

If one views aggregate intake not in terms of total arsenic intake but in terms of toxicologically significant forms of the element, then much of the dietary fraction, for reasons given earlier, such as complex organoarsenicals being present, becomes relatively less important than the forms in water and air as well as in cigarette smoke. Arsenic forms in such media include: pentavalent arsenic in most water supplies; variable mixtures of tri-and pentavalent arsenic in ambient air; and probably an arsenic oxide in cigarette smoke. From this view point, utilizing the examples already given above, nonsmokers would absorb 20 μg or less daily of toxicologically significant arsenic. Heavy smokers having otherwise very low air and water exposure, conceivably could receive their major exposure via cigarettes.

9.2 SIGNIFICANT HUMAN HEALTH EFFECTS ASSOCIATED WITH AMBIENT EXPOSURES

9.2.1 Acute Exposure Effects

Serious acute effects and late sequelae from exposure to arsenic will appear after single or short-term respiratory or oral exposures to large amounts of arsenic. Available data indicate that inorganic trivalent compounds of arsenic are generally more acutely toxic than inorganic pentavalent compounds, which in turn are more toxic than organic arsenic compounds. Serious effects will also appear after long-term exposure to respiratory or oral doses of arsenic.

The acute symptoms following oral exposure consist of gastrointestinal disturbances, which may be so severe that secondary cardiovascular effects and shock may result and cause death. Also, direct toxic effects on the liver, blood-forming organs, the central and peripheral nervous systems, and the cardiovascular system may appear. Some symptoms, especially those from the nervous system, may appear a long time after exposure has ceased and may not be reversible, whereas the other effects seem to be reversible. Infants and young children especially are susceptible with regard to effects on the central nervous system. The Japanese follow-up after the Morinaga milk poisoning showed that persisting damage, especially mental retardation and epilepsy, is a late sequela in children of short-term oral exposure to large doses of inorganic arsenic. Among adults, the central nervous system is not as susceptible, but peripheral neuropathy has been a common finding.

Both in adults and children, acute oral exposure has resulted in dermal changes, especially hyperpigmentation and keratosis, as a late sequela.

Acute inhalation exposures have also resulted in irritation of the upper respiratory tract, even leading to nasal perforations.

Direct dermal exposure to arsenic may lead to dermal changes; allergic reactions may also be involved.

9.2.2 Chronic Exposure Effects

Both carcinogenic and non-carcinogenic effects are associated with long-term exposures, which do not cause any obvious immediate effects. For the purpose of this document, such chronic effects will be discussed in sequence as follows:

1. Respiratory tract cancer
2. Skin cancer
3. Non-cancerous skin lesions
4. Peripheral neuropathological effects
5. Cardiovascular changes

Cancer of the respiratory system is clearly associated with exposure to arsenic via inhalation. This association has been especially noted among workers engaged in the production and usage of pesticides and among smelter workers. While it is not known to what extent exposure to other compounds in industrial atmospheres has contributed to the excess of lung cancer, the Carcinogen Assessment Group (CAG) has concluded that there is sufficient evidence that inorganic arsenic compounds are lung carcinogens in humans.

Cancer of the skin has been found as a dose-related effect in a population in Taiwan, with lifetime exposure to arsenic in well water. It has also been found among people treated with large doses of arsenite for skin disorders. Skin cancer often has a long latency period on the order of decades, the latency time decreasing with increasing intensity of exposure. The CAG has concluded that there is sufficient evidence that inorganic arsenic compounds are skin carcinogens in humans.

Hyperkeratosis and hyperpigmentation, sometimes with precancerous changes, have been a common finding in persons ingesting arsenic. These skin lesions, as well as the manifest cancer, develop on skin surfaces usually unexposed to sunlight. In studies in the United States, an association between skin lesions or skin cancer has not been demonstrated. These studies have been limited, however, by sample sizes too small to be able to detect the dose response seen in studies outside the U.S.

The effects on the peripheral nervous system range from sensory disturbances to motor weakness and even paralysis. The more severe signs have been noted in subacute poisonings, but more subtle changes after long-term low-level exposure have been found by using electromyography or measuring nerve conduction velocity. These subclinical effects are slow in recovery and may persist for years after cessation of exposure. In a study in Canada, electromyographic (EMG) changes were noted when water concentrations of arsenic exceeded 0.05 mg/l.

Cardiovascular effects have been noted especially in Taiwan, where Blackfoot disease (peripheral vasculopathy) occurred after long-term exposure to arsenic in well water. However, the presence of ergotamine-like compounds raises the possibility of vascular effects from these agents. Peripheral vascular changes were also found among German vintners who were exposed both occupationally, by spraying arsenic-containing pesticides, and orally, by drinking wine with elevated arsenic levels. Studies on occupationally-exposed persons have been inconclusive in showing that arsenic causes an increase in mortality from cardiac disease.

9.3 DOSE-EFFECT/DOSE-RESPONSE RELATIONSHIPS

9.3.1 General Considerations

This section generally attempts to define, as presently feasible, human dose-effect/dose-response relationships for health effects of likely greatest concern at ambient environment exposure levels for arsenic in the United States. As such, the present section highlights mainly the quantitative carcinogenic risk estimates that were derived in Section 7.3.

The general question of how to define and employ a dose factor in attempts at quantitative assessments of human health risk for any toxicant is highly dependent upon: 1) the available information on the body's ability to metabolize the agent, and 2) the assessment of the relative utility of various internal indices of exposure.

The time period over which a given total intake occurs is highly important. For example, intake of one gram of arsenic over a period of years would be quite different pathophysiologically from assimilating this amount at one time, the latter probably having a lethal outcome. This time-dependent behavior is related in part to the relative ability of the body to detoxify inorganic arsenic by methylation as a function of both dose and time.

In cases of acute and sub-acute exposure, indicators of internal exposure such as blood or urine arsenic levels are probably appropriate for assessing the intensity of exposure.

With chronic, low-level exposure, however, the available data would indicate that the total amount assimilated is probably more important than an indicator concentration without knowledge of the total exposure period. An added problem is the background level of arsenic found in these indicators due to dietary habits. For example, in acute exposures, levels in blood or urine would be greatly elevated over background values while low-level chronic exposures would only result in moderate increases over background.

In regard to hair arsenic levels as an indicator of internal arsenic exposure, no reliable methods exist for distinguishing external contamination levels from those accumulated via absorption and metabolic distribution. Hair arsenic levels cannot, therefore, be employed as reliable indicators of either current or cumulative long-term exposures for individual subjects, but rather may provide only a rough overall indication of group exposure situations.

Given the above considerations and limitations concerning the use of blood, urinary or hair arsenic concentrations as internal indices of cumulative, long-term low-level arsenic exposures of concern here, the dose-effect/dose-response relationships summarized below are done so mainly in terms of external arsenic exposure levels via either inhalation or ingestion.

9.3.2 Effects and Dose-Response Relationships

It is difficult to define a precise acute lethal dose of arsenic for man, because such exposure situations rarely allow accurate determination of the effective amounts. However, for trivalent arsenic, the figure is believed to range from 70 - 180 milligrams.

For subacute exposure, it appears that for children, about one gram assimilated over a period of 3-4 weeks will induce death with severe effects in survivors, while for adults, that dose will occasion significant clinical effects. In one poisoning episode, intake of approximately 50 milligrams over a period as short as two weeks resulted in clinically demonstrable effects in adults.

9.3.2.1 Respiratory Cancer--A considerable number of studies have shown associations between occupational exposure to arsenic and cancer of the respiratory system. The best information available for making quantitative risk estimates for lung cancer are derived from 5 sets of data involving 4 sets of investigators and 2 distinct exposed populations. The 4 sets of investigators are Brown and Chu (1983a,b,c), Lee-Feldstein (1983) and Higgins (1982)--who conducted studies on workers at the Anaconda smelter in Montana--and Enterline and Marsh (1982)--who conducted a study on the workers at the ASARCO smelter in Tacoma, Washington.

Using an absolute-risk linear model and the data from the four smelter studies, the lifetime lung cancer risk, due to continuous exposure of 1 μg/As/m^3, was estimated to range from 1.25 x 10^{-3} to 7.6 x 10^{-3}. A weighted average of the five estimates* in this range gave a composite estimate of 4.29 x 10^{-3} (see Section 7.3.2.3). This represents a plausible estimate of the upper limit of risk--that is, the true risk would not likely be more than the estimated risk, but it could be substantially lower.

9.3.2.2 Skin Cancer--Chronic arsenic exposure, both occupational and non-occupational, is associated with a distinctive hyperkeratosis, which is usually followed by a later onset of skin cancer. The best data available for making quantitative risk estimates for skin cancer are the data collected by Tseng et al. (1968). In this study, the authors surveyed a stable population of 40,421 individuals who lived in a rural area along the southwest coast of Taiwan and who were known to have consumed drinking water containing arsenic. The occurrences of skin cancer among this population and the arsenic concentrations in their drinking water were measured. Since the population was stable, the data obtained from the study lends itself to predictions of lifetime probability of skin cancer caused by the ingestion of arsenic.

Using an absolute-risk linear model and the data from Tseng et al., the lifetime skin cancer risk from drinking water containing 1 μg/liter of arsenic was estimated to be 4.3 x 10^{-4} (see Section 7.3.3). It is not likely that the true risk for skin cancer would be more than this estimated risk, but it could be considerably lower.

9.3.2.3 Non-cancerous Skin Lesions -- As noted above, in man, chronic oral exposure to arsenic induces a sequence of changes in skin epithelium, proceeding from hyperpigmentation to hyperkeratosis, characterized as keratin proliferation of a verrucose nature and leading, in some cases, to late onset skin cancers. These effects have been noted both in populations which have ingested arsenic via drinking water and among people treated with large doses of arsenite for skin disorders. In a report by Pershagen and Vahter (1979), the authors, using the data from a patient population exposed to arsenic via Fowler's solution (Fierz, 1965), noted an increase in prevalence of hyperkeratosis with increasing dose of arsenic. The U.S. EPA is presently examining this information, along with information from other studies, in order to determine whether quantitative dose-response relationships, similar to those seen for skin cancer, can be established for these precancerous skin lesions.

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*Two risk estimates were derived from the Enterline and Marsh study based upon exposure periods lagged 0 and 10 years. See Table 7-25.

9.3.2.4 Peripheral Neuropathological Effects and Cardiovascular Changes -- While the qualitative evidence for peripheral neurological effects and cardiovascular changes in arsenic-exposed populations is incontrovertible, the data are insufficient to establish quantitative dose-response relationships at the present time.

9.4 POPULATIONS AT SPECIAL RISK TO ARSENIC EXPOSURE

In reviewing the literature dealing with the acute, subacute and chronic effects of arsenic in children and adults, the evidence suggests that children may be at special risk for the effects of inorganic arsenic under conditions of acute or subacute exposure.

In earlier sections, reference was made to the outbreak of pediatric poisoning by arsenic in Japan due to the presence of arsenic in infant milk formula (Hamamoto et al., 1955). From the clinical reports published at the time of the mass poisoning as well as those from follow-up studies, a number of signs of central nervous system involvement were noted both at the time of the episode and much later, with the follow-up studies showing behavioral problems, abnormal brain wave patterns, marked cognitive deficits, and severe hearing loss in some of those children who survived the acute episode. Some of these same tardive effects have also been noted in adults but appear to be a much less constant feature of arsenic-induced neurotoxic effects than are the peripheral neuropathies.

Because children consume more water per body weight than do adults, the daily intake of arsenic via drinking water per kilogram body weight would be greater in children. This may have implications regarding chronic exposure effects in children. Zaldivar (1977) developed a regression equation describing this relationship. It should be noted, however, that serious health effects due to chronic exposure of arsenic in drinking water have not been found at a greater frequency in children than adults.

Individuals residing in the vicinity of certain arsenic-emitting sources, e.g., certain types of smelters, may be at risk for increased arsenic intake because of both direct exposure to arsenic in air and indirect exposure via arsenic secondarily deposited from air onto soil or other human exposure media. The relative contribution of such indirect exposures to increased risk of these individuals for arsenic health effects is difficult to define due to the lack of information on this subject. However, it is most likely minimal in relation to the direct effects arising from inhalation of arsenic, including lung cancer.

As a large class of the general population at risk for increased arsenic intake, one would have to include cigarette smokers. However, it is not clear to what extent some increased arsenic intake from tobacco smoke poses a specific heightened health effect risk, although it is clear that internal indicator levels, e.g. blood arsenic, are somewhat elevated in the case of cigarette smokers relative to nonsmokers.

10. REFERENCES

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